The dietary patterns, socio-demographic and lifestyle determinants, and metabolite fingerprints in a community sample of Merseyside takeaway food consumers

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ABSTRACT

Background: Takeaway foods have increased in popularity over the past 40 years and there has been much debate regarding the determinants of their consumption. Whilst takeaway foods are thought to be associated with a number of non-communicable diseases, wider dietary patterns may be more predictive of disease risk than individual foods due to the synergistic effects of food combinations. However, accurate measurement of habitual food, particularly energy-dense nutrient-poor foods, using self-reported dietary assessment tools can be subject to participant bias. Recent research has demonstrated that metabolites derived from individual foods present in urine samples provide biomarkers of dietary exposure. ***Aim of PhD:*** To investigate takeaway food consumption within wider dietary patterns of a Merseyside population and their socio-demographic and lifestyle determinants. Additionally, to identify novel metabolite biomarkers from urine samples associated with takeaway foods as a means of validation of self-reported food consumption. ***Methods:*** Study 1) Design a food frequency questionnaire (Eating Habits Survey (EHS)) and conduct pilot (n = 26) to test the efficacy of methods prior to a larger scale study. Study 2) Record habitual and takeaway food consumption of study population (n = 1724) using the EHS and use principal component analysis (PCA) to determine dietary patterns. Study 3) Validate the EHS using three 24 hour 'Multiple Pass' dietary recalls (3 x 24 h MPR) and metabolomics on urine samples from a sub-sample (n = 151) from Study 2. ***Results:*** Being male, in a younger age group, having children in the household, alcohol use and smoking were positively associated ($p < 0.05$) with takeaway food consumption, whereas being in an older age group and having higher qualifications were negatively ($p < 0.01$) associated. Nevertheless, specific takeaway food groups were associated at varying degrees. PCA identified some contrasting dietary patterns from the EHS and 3 x 24 h MPR, many of which included ultra-processed foods, and two concurring major patterns; ‘Western’ and ‘Prudent’. Metabolomics determined, for the first time, differences among the metabolite fingerprints of takeaway food consumers, and has shown good biomarker potential for kebab, Indian, English and Chinese takeaway food. Metabolomics validated self-reported dietary intake and determined urinary metabolites associated with habitual exposure to specific foods including poultry, coffee, alcohol, cocoa, oils, and fruit and vegetables. ***Conclusion:*** This research makes a novel contribution to knowledge and offers valuable insight into the determinants of takeaway food consumption and wider dietary patterns in the UK adult population. Moreover, it has built on an innovative metabolite fingerprinting technique that distinguishes food exposure from urinary samples. These findings should inform further research in this highly topical area and provide the evidence base to influence the formation of policy and interventions on takeaway food locally and in other areas.

Key words: Dietary assessment, dietary exposure, dietary survey, fast food, food frequency questionnaire, metabolomics, multiple pass dietary recall, nutritional biomarkers, obesity, overweight, out-of-home foods, principal component analysis, prudent, western, takeout.

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Glossary of terms

24 h MPR	**24 hour 'Multiple Pass' dietary recall**
ANOVA	**Analysis of Variance**
AU	**Aberystwyth University**
AUC	**Area under the curve**
BMI	**Body Mass Index**
CEDAR	**Centre for Diet and Activity Research**
CVD	**Cardiovascular disease**
DF	**Discriminant Functions**
EHS	**Eating Habits Survey**
EPIC	**European Prospective Investigation of Cancer**
FFQ	**Food frequency questionnaire**
FIE-HRMS	**Flow Infusion-High Resolution Fingerprinting**
HTA	**Human Tissue Authority**
IBERS	**Institute of Biological, Environmental & Rural Sciences**
IMD	**Index of multiple deprivation**
IQR	**Interquartile range**
LJMU	**Liverpool John Moores University**
LPA	**Low physical activity**
M	**Mean**
MANOVA	**Multivariate Analysis of Variance**
MED	**Median**
MS	**Mass spectrometry**
MTA	**Materials transfer agreement**

MVPA Moderate and vigorous physical activity

NDNS National Diet and Nutrition Survey

NS-SEC National Statistics Socio-economic Classifications

PA Physical activity

PCA Principal component analysis

PC-LDA Principal component–linear discriminant analysis

PHE Public Health England

RF Random Forest

SD Standard deviation

SSB Sugar-sweetened beverages

UHPLC-HR Ultra High Performance Liquid Chromatography-High Resolution

UK United Kingdom

US/A United States of America

WHO World Health Organization

WHtR Waist to height ratio

Chapter 1
Introduction

1 CHAPTER 1: INTRODUCTION

1.1 ORGANISATION OF THESIS

The overall theme of the thesis is investigating takeaway food consumption, wider dietary patterns, determinants of consumption and biomarkers associated with specific food intakes in a sample of takeaway food consumers. Previous research has investigated the composition of takeaways from independent takeaway food outlets, however, gaps were identified in the literature regarding the dietary significance of such foods and how takeaway foods fit within wider dietary patterns of food consumption. In addition, nutrition research often describes the challenge of generating accurate dietary information using traditional dietary assessment techniques and has evidenced new and emerging techniques such as metabolomics as a means of validating self-reported dietary consumption. This research has for the first time, aimed to fill these research gaps by making a contribution to knowledge on takeaway food consumers along with the novel use of urine biomarkers to validate findings and identify potentially new biomarkers associated with takeaway food exposure. **Chapter 1** introduces the project and describes the rationale for studying takeaway food consumption in the UK. This chapter provides an overview of takeaway foods, the link to obesity, cardiometabolic risk etc., the takeaway food environment and health in Merseyside, and previous research conducted at LJMU on the nutritional composition of takeaway foods in Merseyside. **Chapter 2** presents a review of the literature. The key themes discussed are takeaway foods and health, factors influencing takeaway food consumption, dietary patterns as a means of dietary assessment, and urinary metabolomics as an objective measure of dietary exposure. **Chapter 3** defines the methodologies used in the study. **Chapter 4, 5 and 6** report the results from three studies. A pilot study (Study 1) to test methods and check the feasibility of the study design. A cross-sectional study (Study 2) to investigate takeaway food intake and wider dietary patterns in a Merseyside community sample and socio-demographic and lifestyle determinants. Lastly, a metabolomics study (Study 3) to explore specific dietary intake and metabolite fingerprints in a sub-sample of takeaway food consumers from Study 2, and to identify known or to discover new chemicals (metabolite biomarkers) and clusters associated with the dietary patterns consumed. **Chapter 7** conveys a synthesis of the findings from all three studies, offers recommendations for future research and draws up an overall conclusion to the project.

1.2 TAKEAWAY FOODS

Takeaway, take out, and fast foods are common terminology used for various 'out-of-home' foods. The term 'takeaway foods', commonly used in the United Kingdom (UK) and Australia, defines hot meals made to order and take away from small, independent outlets (Miura et al., 2012; Jaworowska et al., 2014); the United States of America (US) 'take-out' shares a similar definition. 'Fast food' mainly defines foods from national/multinational fast food chains (such as McDonald's, Domino's Pizza, Subway, Burger King, Pizza Hut, Kentucky Fried Chicken and Taco Bell) (Block et al., 2004; Richardson et al., 2011; Bauer et al., 2012), and can include dining in. However, 'out-of-home' foods do include multiple definitions and can come from a number of sources including vending machines, convenience stores, fast food outlets, takeaway food outlets, coffee shops, schools, etc. (Nago et al., 2014). For the purpose of this PhD project, the terminology from the original articles reviewed has been maintained to represent the subtle differences between studies. Where multiple studies are being discussed and in instances of critique, ‘takeaway foods’ has been used as this term broadly covers takeaway and fast food.

Out-of-home foods have become increasingly popular over the past few decades and are thought to be one of the key components driving increasing levels of overweight and obese individuals (Lachat et al., 2012). The causes of obesity are complex (Butland et al., 2007) but the overconsumption of food and sugar-sweetened beverages (SSB), along with increased portion sizes, are also undoubtedly strong determinants (Marteau et al., 2015). A recent UK study found that 27% of adults and 19% of children consumed meals outside the home once per week or more and 21% of adults and children ate takeaway meals at home once per week or more (Adams et al., 2015). Similar consumption patterns are common in other high income and urban societies; particularly those in Europe, the US and Australia (Guthrie et al., 2002; Orfanos et al., 2007). Kant et al. (2015) found that more than 50% of US adults reported consuming 3 or more out-of-home meals a week and more than 35% reported consuming 2 or more fast food meals per week. While Australia, the UK, and the US have shaped the majority of research on takeaways. The same issues (poor dietary habits and increased prevalence of non-communicable disease) are of equal concern for urban centres in developing economies undergoing ‘nutrition transition’ (Popkin and Gordon-Larsen, 2004); these include Asia, Africa, the Middle East and Latin America.

Out-of-home foods tend to be less healthy, because they are more energy-dense and nutrient-poor, than foods prepared at home (Lachat et al., 2012;

Nguyen and Powell, 2014). They often contain high quantities of fat, salt and sugar, which are associated with weight gain and a variety of negative health outcomes (Duffey et al., 2009; Jaworowska et al., 2012; Jaworowska et al., 2014). Frequent consumption of fast food and takeaway food has been associated with higher body mass index (BMI) and biomarkers of greater cardiometabolic risk (Duffey et al., 2009; Smith et al., 2012; Kant et al., 2015). While there is a consensus that being overweight (BMI 25–29.9 kg/m^2) and obese (BMI ≥ 30 kg/m^2) (National Institute of Health and Care Excellence, 2014; World Health Organization, 2016) is associated with high consumption of energy-dense and nutrient-poor foods, the factors influencing their consumption are not well understood. Furthermore, there is no single causative factor to becoming overweight or obese although unhealthy dietary patterns are considered a key factor that warrant intense investigation (Butland et al., 2007).

1.3 Obesity

Over the past few decades, the development of convenient fast food and takeaway food has competed successfully against home-prepared food in Western societies (Guthrie et al., 2002; Trapp et al., 2015). Concurrently, obesity has been spreading like an epidemic (Mitchell et al., 2011). This has become a global issue where more than one in three adults are overweight or obese (World Health Organization, 2018). Energy-dense nutrient-poor diets are significantly associated with overweight and obesity and also contribute directly to a variety of other causes of disability and death (Adams et al., 2016). The prevalence of obesity among adults in England has almost doubled from 15% in 1993 to 26%

in 2016/17 but has remained at a similar level since 2010 (National Health Service Digital, 2018). The shifting from healthy weight to obese based on BMI has created a bell-curve in populations characterised by a sharp increase in the obese categories and little or no change in the overweight category (Bray, 2015).

Takeaway food is regularly referred to when speaking about the obesity epidemic due to the association with higher intakes of sugar and fat and larger portion sizes (Brindal et al., 2008). A large scale study by Smith et al. (2013) showed a shift in dietary patterns and food preparation since 1965 due to a significant decline in time spent cooking in the home and growing trends in out-of-home food consumption. A UK study aiming to document the prevalence of time spent cooking in 2005 showed that 60% of women and 33% of men reported spending 30 minutes of continuous cooking daily (Adams and White, 2015). Less time spent cooking could be an indicator of increased consumption of takeaway foods. The findings suggested that being female was the main determinant of time spent cooking, with little influence from older age, greater education, unemployment, lower social class and living with others (Adams and White, 2015). Nevertheless, the level of attrition within the study was substantial and data collected in 2005 may not represent accurately more recent trends.

The shift in out-of-home food choice coupled with an increase in sedentary behaviour has contributed to an obesity epidemic in the 21st Century (Jacobs, 2006; Lachat et al., 2012; Pieroni and Salmasi, 2014). Lowry et al. (2015) found a positive association between television/computer screen time and consumption of fast food and SSB in a sample of students (Appendix 9.1). The findings suggested a pattern of unhealthy behaviours which support previous research stating that television viewing and fast food consumption was positively associated with BMI (Jeffery and French, 1998). The use of takeaway and out-of-home foods may also be attributable to individuals working more and experiencing feelings of time scarcity; this has been especially evident among women (French et al., 2001; Jabs et al., 2007; Welch et al., 2009) (Appendix 9.1). Urbanisation, economic growth and educational achievement in low- and middle-income countries have all been shown to influence the consumption of energy-dense nutrient poor foods (Malik et al., 2013). One study presented findings that individuals seeking professional success wanted to avoid spending time and effort clearing up after meals, to create time for other activities (Botonaki and Mattas, 2010) (Appendix 9.1). As a result, it would appear that the time constraints of working long hours coupled with the advances of new technology may contribute to an increase in people's consumption of out-of-home energy dense foods (Pieroni and Salmasi, 2014).

1.4 MERSEYSIDE FOOD AND HEALTH ENVIRONMENT

Merseyside is a county in the North West of England with a total population of 1,406,400 (685,600 males, 720,900 females) and 893,000 (439,800 males, 453,200 females) aged 16–64 years (ONS Population estimates, 2016). Merseyside is made up of 5 districts; Liverpool, Metropolitan Borough of Sefton, Metropolitan Borough of Knowsley, Metropolitan Borough of St. Helens, and Metropolitan Borough of Wirral (Fig. 1.1). According to the most recent statistics on obesity, the North West had the third highest prevalence of obesity in England, at 30% of the population (National Health Service, 2017).

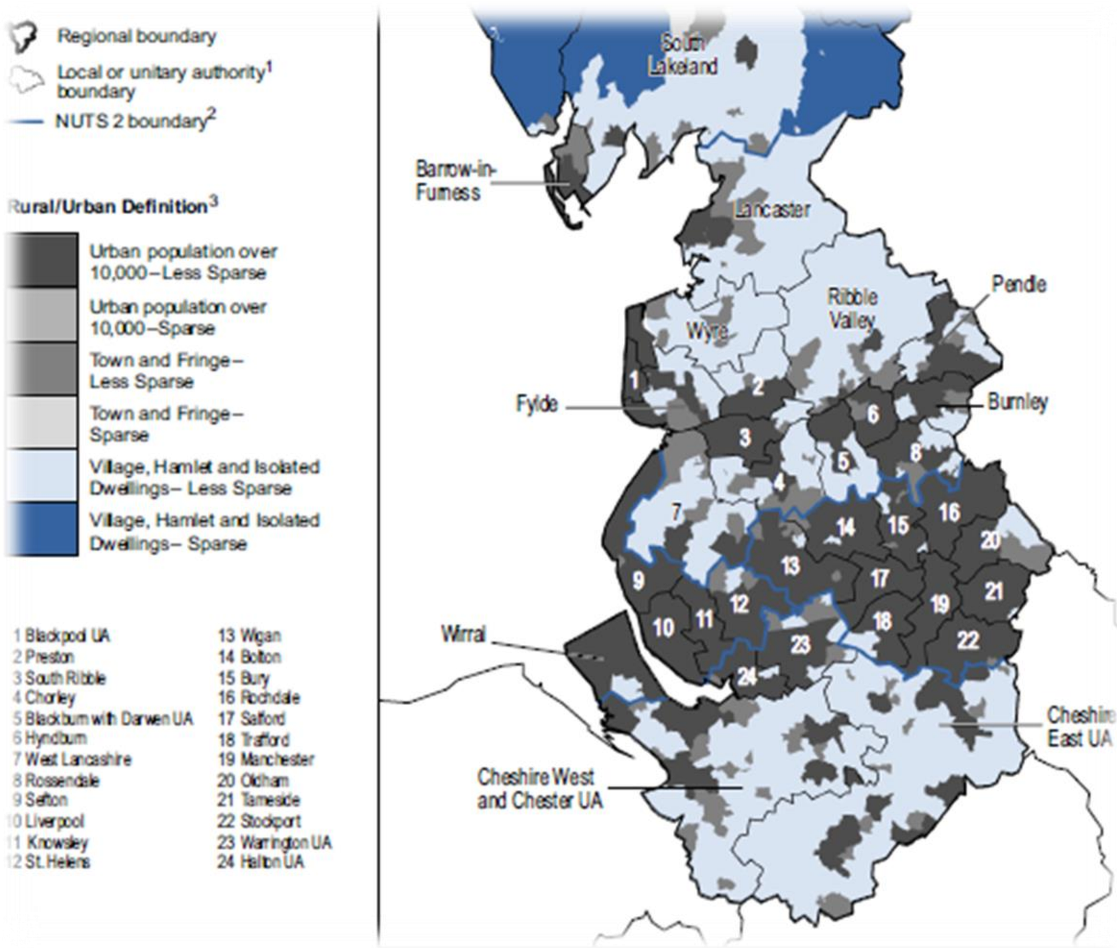


Figure 1-1 Districts 9–12 and the Wirral contribute to Merseyside County (Office for National Statistics, 2011)

Data from 2015/16 showed that the prevalence of overweight (and obese) adults aged 18+ in Merseyside was 65.7% (ranging from 59.6% in the Wirral to 72.1% in St. Helens) worse than the England average of 61.3% (Public Health England, 2018c). Public Health England (2018c) also reported that only 49.9% of adults in Merseyside meet the recommended ‘5-a-day’ on a ‘usual day’ for fruit and vegetable intake. The average number of portions of fruit and vegetables consumed was 2.44 respectively. A selection of population characteristics are detailed below (Table 1.1). Based on the statistics from Public Health England

Merseyside appears to have substantial health inequality compared to the England average.

Table 1-1 Characteristics of the Merseyside population (Public Health England, 2018c)

Characteristic	Merseyside average	England average
Overweight and obese adults	65.7%	61.3%
Diabetes (recorded cases aged > 17)	6.6% (7.1% in Knowsley and St. Helens)	6.4%
Cardiovascular disease mortality rates for adults < 75 years	89 per 100,000	73.5 per 100,000
Physically active adults > 19 years	63.2%	64.9%
Current smokers	16.9% (12.8% in Sefton)	15.5%
Alcohol related hospital admissions	856 per 100,000	636 per 100,000
Index of multiple deprivation (IMD) score: lower score being favourable	32.92 IMD (ranging from 25.7 Sefton to 41.4 Knowsley)	21.8 IMD

Merseyside is a relatively small county compared to others in North West England but is densely populated and has some of the highest densities of takeaway food outlets in the country. Food outlet data from September 2017 (UKCRC Centre for Diet and Activity Research (CEDAR), 2017b) showed that 28.1%–34.5% (n=1633) of the total number of food outlet types in Merseyside districts were takeaways, higher than the England average of ~25% (n=56,638). Furthermore, according to CEDAR (2017b) Liverpool had one of the highest proportions of takeaway food outlets per resident population in the country (1.44/1K population). However, when looking at middle super output areas these results varied considerably across Merseyside (Fig. 1.2).

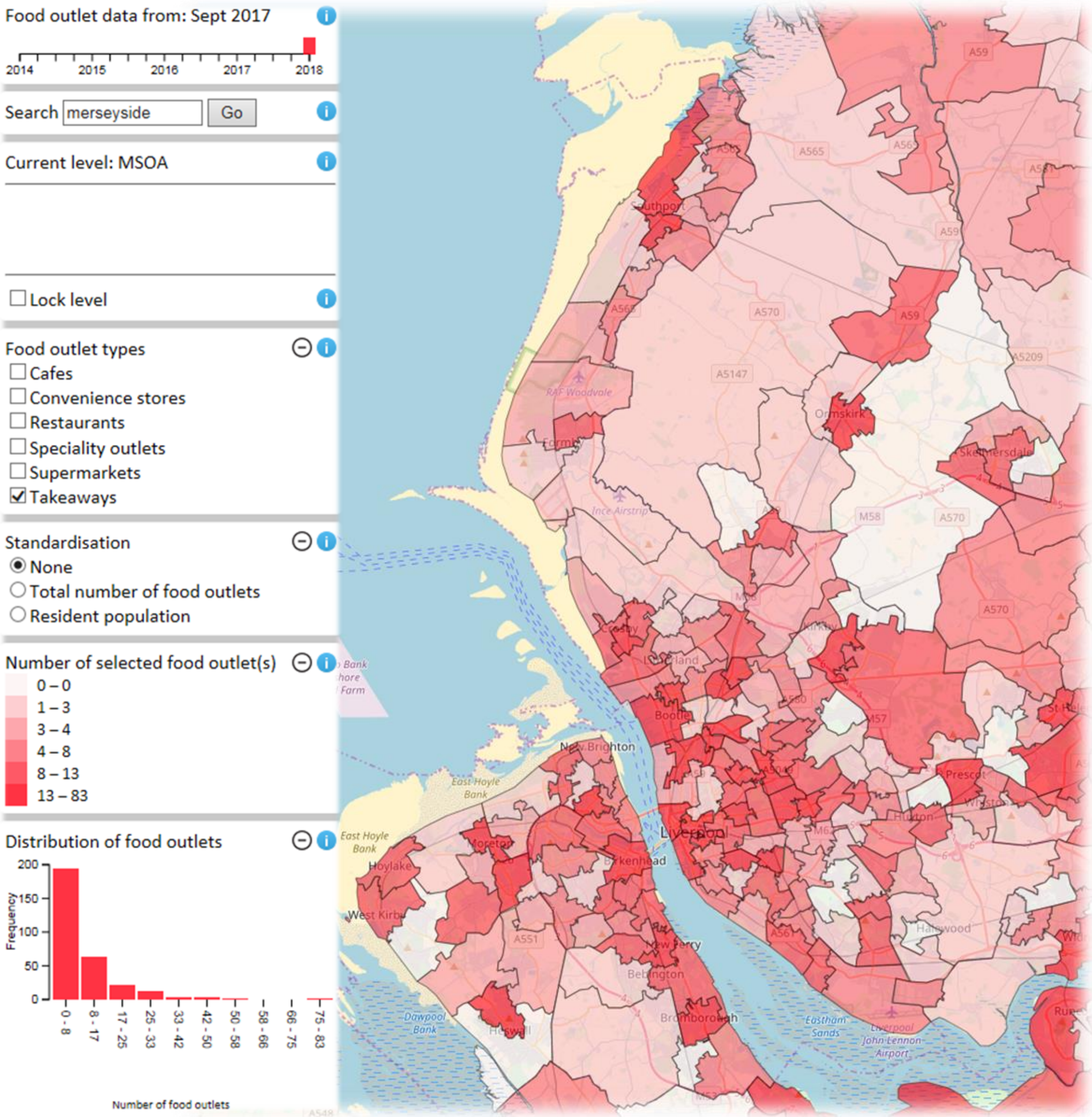


Figure 1-2 Number of takeaway food outlets in middle super output areas (UKCRC Centre for Diet and Activity Research (CEDAR), 2017b)

1.5 Eatright Liverpool

A previous project undertaken by researchers at LJMU, on the nutritional composition of takeaway food from small independent takeaway outlets in Merseyside, demonstrated potential nutritional problems with many of the meals sampled, particularly their excessive energy; fat; sugar; and salt levels (Jaworowska et al., 2014). The project also established recipe reformulations and changes to food preparation and cooking practices, as a means of improving the nutritional composition of the takeaway food (Jaworowska et al., 2011). Nonetheless, focussing on the nutritional composition of individual meals limited the understanding of the dietary impact to health due to the absence of additional data on consumption (frequency, portion size and additional foods). Hence, the need for further dietary survey work. Moreover, research over the last decade has shifted towards analysing dietary patterns as a more global approach to examining diet-disease related relationships (McNaughton 2010).

1.6 RATIONALE

Previous research at LJMU has indicated that the nutritional composition of takeaway food is potentially unfavourable to health and it is widely acknowledged that takeaway food consumption is linked to increasing obesity levels and non-communicable disease. Merseyside is a hotspot for takeaway outlets and has a high prevalence of obesity and poor health outcomes, which makes it an ideal location to analyse takeaway food consumption in more detail as a primary step towards supporting future nutrition/health policies and interventions. Dietary impact from takeaway food is somewhat driven by the composition of the food(s), ranging from healthy to unhealthy options, but also dependant on a number of other variables including frequency of feeding, food choice, habitual diet, and combinations of food intake (overall dietary pattern). Therefore, it was necessary to explore takeaway food intake in detail alongside habitual food intake.

Data from the most widely referred to UK National Diet and Nutrition Survey (NDNS) has shown an association between eating out-of-home foods and less healthy food choices in adults (Ziauddeen et al., 2017). However, the NDNS has limited information regarding types of takeaway foods consumed, frequency of consumption and portion sizes (Ziauddeen et al., 2017). Therefore, as recommended by Subar et al. (2015) the current study was designed to collect dietary data on habitual and takeaway foods from both short-term (24 hour 'Multiple Pass' dietary recalls (24 h MPR)) and long-term (food frequency questionnaire (FFQ)) techniques to allow for maximizing the strengths of each technique. The inclusion of dietary biomarkers via a new metabolite fingerprinting technology provided an opportunity to enhance these dietary assessment tools.

Individuals and populations consume foods, not nutrients, and foods are consumed synergistically as part of an overall diet and lifestyle (UKCRC Centre for Diet and Activity Research (CEDAR), 2017a). Instead of focusing on single nutrients, dietary pattern analysis studies the intake of foods and nutrients in combinations and may be more predictive of health outcomes. It was especially important to investigate the dietary patterns of takeaway consumers to understand how takeaway food was consumed in relation to habitual foods. Understanding the complexities of these dietary patterns and what factors may influence them is important for identifying groups at greater risk of obesity and non-communicable disease, and could help to inform strategies and policies to improve population diet and health.

Food choices and eating behaviours are influenced by many interrelated factors including cultural, environmental, demographic, biological, cognitive and behavioural (Story et al., 2008; Franchi, 2012); hence the need for a holistic approach. Furthermore, in order to create effective interventions that can be delivered at the population level to improve health, irrespective of risk (Rose, 1992; Rose, 2001) evidence regarding food consumption, and information on socio-demographics and lifestyle determinants are warranted to shape such interventions.

1.7 AIM AND OBJECTIVES

This study aimed to investigate takeaway food consumption within wider dietary patterns of a Merseyside population and their socio-demographic and lifestyle determinants. Additionally, this study aimed to identify novel chemical (metabolite biomarker) clusters, which are associated with takeaway foods/unhealthy eating as a means of validation of self-reported food consumption and to add to a biomarker panel/dietary exposure tool for future applied human nutrition research. Overall, this study aimed to make a contribution to knowledge regarding takeaway food consumers, dietary patterns derived through principal component analysis (PCA) and an innovative dietary assessment technique using urine samples to determine food exposure.

Objectives:

1. To develop and trial a modified FFQ on a pilot sample prior to conducting a larger cross-sectional study to assess habitual and takeaway food intake and identify dietary patterns in relation to socio-demographic, anthropometric and lifestyle factors.

2. To identify dietary patterns of the pilot sample from three 24 hour 'Multiple Pass' dietary recalls (3 x 24 h MPR) to evaluate differences between two dietary assessment methods and as a validation technique for the modified FFQ.
3. To explain the habitual and takeaway food consumption, socio-demographic and lifestyle characteristics and wider dietary patterns of a cross-sectional Merseyside population.
4. To determine the dietary patterns of a sub-sample of data-driven takeaway food consumer groups (high, medium, and low) and validate findings using two forms of self-reported dietary assessment and one biomarker technique.
5. To classify metabolite fingerprints in urine indicative of takeaway foods/unhealthy foods and to add to a model for objective classification of dietary exposure based on urinary metabolite fingerprint data.

Chapter 2
Literature review

2 Chapter 2: Literature Review

2.1 Introduction

This literature review starts by setting the scene of the current research on takeaway food consumers and the associated determinants of takeaway food consumption, highlighting areas of conflicting evidence. The review then evolves into a more focussed critique of dietary pattern analysis and metabolomics analysis as new and emerging methods of dietary assessment. Specifically how to tailor such techniques for takeaway food consumers, whilst focusing on the research gaps. Research has shown a diversity in nutritional composition, both between and within types of takeaway food (Jaworowska et al., 2014). Thus, it may be assumed that not all self-reported frequent consumers of takeaway food eat the same and may produce contrasting dietary patterns. In spite of the evidence from around the globe, there is a current gap in the literature concerning takeaway food consumption in relation to wider dietary patterns in UK adults. Dietary patterns provide an holistic way of explaining population food intake that includes a comprehensive assessment of the diet. Dietary pattern analysis of takeaway food consumers may provide insights into the subtle differences between their habitual and takeaway food consumption and may be more predictive of poor health outcomes than frequency of consumption. It is also important to consider the socio-demographic and lifestyle determinants to explain reasons for these subtle differences. A number of limitations such as recall bias, memory loss, and inaccurate measurements constantly challenge the validity and accuracy of dietary assessment techniques. Moreover, there have been limited targeted techniques for analysing takeaway food consumption. Therefore, to complement the ‘subjective’ traditional dietary assessment techniques, this study aimed to include a novel ‘objective’ method of producing metabolite fingerprints that may be indicative of food intake.

Diet is one of the main factors in the maintenance of good health and wellbeing and plays a significant role in the prevention of many non-communicable diseases (World Health Organization, 2010). Yet, many of the environments that populations now live in are not conducive to the consumption of healthy food but more so for the consumption of energy-dense nutrient-poor out-of-home foods. According to CEDAR (2017b), the number of takeaway food outlets in England is increasing, with an estimated +57,000 outlets in 2014. An Office for National Statistics report (2018) evidenced increasing numbers of takeaway outlets on UK high streets; from 47 per 100,000 people in 2010, to 61 in 2018. Previous research shows the association between increased access to takeaway food outlets and obesity (Burgoine et al., 2016) suggesting that the proliferation of takeaway food outlets creates less overall choice for healthier options. There are takeaway food studies sourced from around the globe. However, a limited amount of targeted research has been conducted in the North West of England, an area with some of the highest obesity rates, other health issues (cardiovascular disease (CVD) and caridometabolic risk) and numbers of takeaway food outlets (Public Health England, 2018b). Individual food choices and eating behaviours are influenced by many interrelated factors including cultural, environmental, demographic, biological, cognitive and behavioural (Story et al., 2008; Franchi, 2012) (Fig. 2.1) and thus requires an intense and collective investigation.

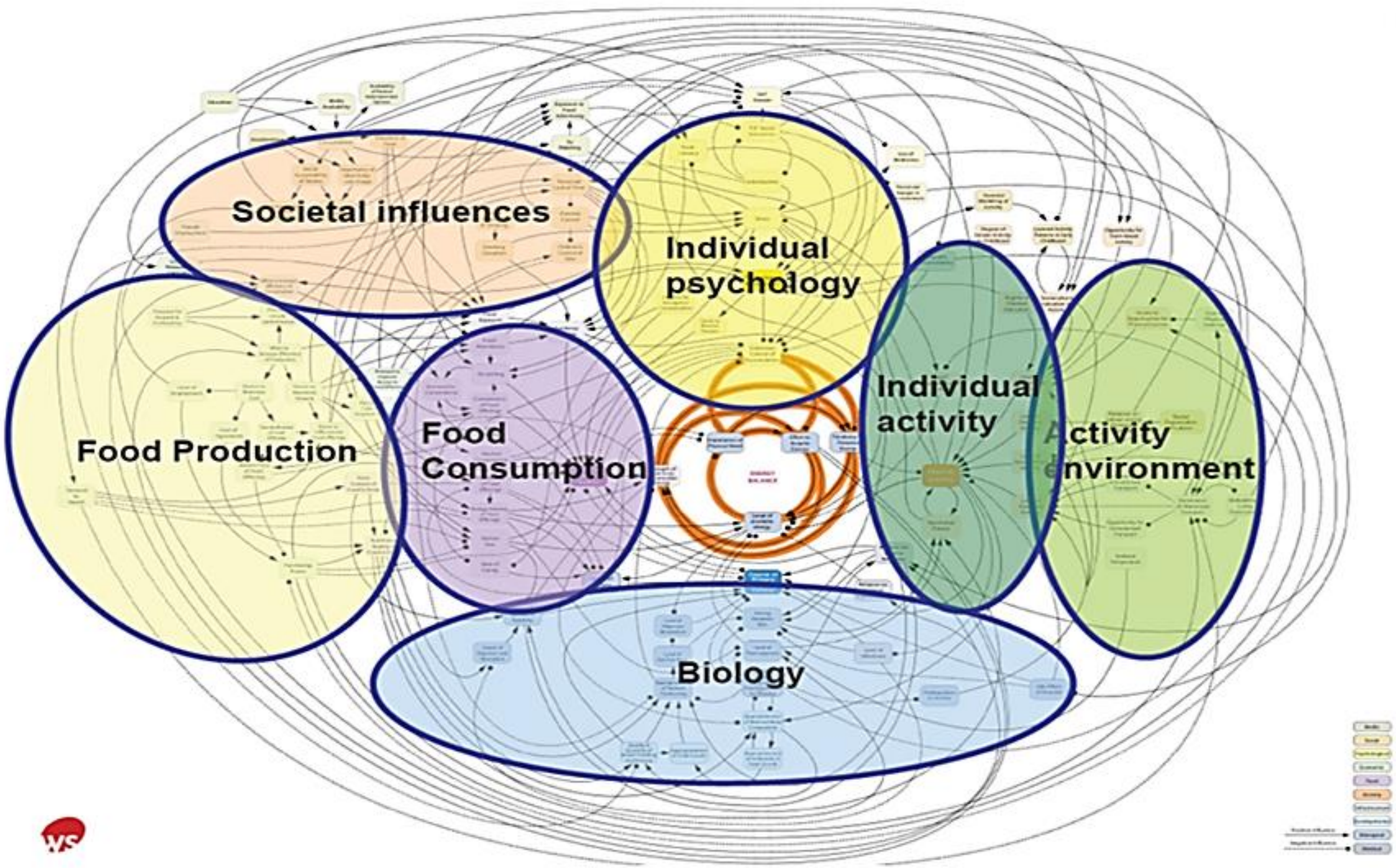


Figure 2-1 Foresight Obesity System Map with thematic clusters (Butland et al., 2007)

2.2 TAKEAWAY FOOD AND GLOBAL HEALTH

Numerous studies have shown increasing trends in frequency of takeaway food consumption, predominately in Europe, US and Australia (Guthrie et al., 2002; Orfanos et al., 2007). Yet, emerging research from low- and middle-income countries including Brazil (Monteiro et al., 2011), Chile (Albala et al., 2002), India (Singh et al., 2015), Iran (Bahadoran et al., 2012), Malaysia (Fournier et al., 2016), Kenya and Tanzania (Keding, 2016) among others, have presented similar findings. Transition to a 'Western diet' is linked with increasing overconsumption of sweets, desserts, soft drinks, red meat, processed meats and high fat dairy products, with a lower consumption of oily fish (omega-3 fatty acids), wholegrains, fruit and vegetables (Hu et al., 2000). Western type diet patterns have become deeply embedded within many societies and despite pressing health related issues continue to grow (Popkin, 2006; Jaworowska et al., 2013). In the UK, a government report using cross-sectional data indicated that 75% of respondents had consumed out-of-home food during the past seven days and 27% had consumed takeaway food (Food Standards Agency, 2014). Figures for takeaway food consumption had increased by 6% from 2010 (Food Standards Agency, 2014). When analysed longitudinally, time devoted to eating and drinking away from home increased significantly in the UK between 1975 and 2000 (Cheng et al., 2007), which concords with the increased prevalence of out-of-home eating establishments seen in parts of the UK between 1980 and 2000 (Burgoine et al., 2009). Similarly, a US study revealed fast food consumption in children increased by 300% between the period of 1977 and 1996 (Fraser et al., 2010). Times of relative scarcity have receded into an era of availability and although most western societies have managed to successfully reduce the burden of infectious disease, the current environment promotes a whole spectrum of dietary induced diseases (Manzel et al., 2014). That said, low- and middle-income countries with existing undernutrition and infectious diseases, that are undergoing development, urbanization and nutrition transition, are now also experiencing a double burden of non-communicable diseases (Agyei-Mensah and de-Graft Aikins, 2010; Bygbjerg, 2012; Shetty, 2013). Therefore, highlighting the urgency of research on out-of-home foods.

Diet is a modifiable determinant of health; however, societies portraying a "Westernised" lifestyle are consuming diets high in out-of-home foods and experiencing a prevalence of obesity (Duffey et al., 2009; Anderson et al., 2015). The challenges in considering the effects of takeaway foods are not solely related to the nutritional composition, but are also dependent on expanding portion sizes (Young and Nestle, 2002). Poor diet and obesity in turn predisposes humans to CVD (Mente, 2009; Carrera-Bastos et al., 2011; Smith et al., 2012), type 2 diabetes (van Dam, 2002; Pereira et al., 2005) and various cancers (Anderson et al., 2015). Interestingly, obesity rates can vary substantially between nations. Although England had an obesity prevalence of 26.8% in 2015; neighbouring European countries demonstrated lower rates such as Germany 23.6% (2012), Belgium 19.2% (2014), and France 17.1% (2015) (Organisation for Economic Co-operation and Development, 2017); suggesting that cultural differences could be a contributing factor. Urban and rural communities in developing economies have also shown contrasting dietary patterns and consequent obesity (Popkin et al., 2012).

2.3 Takeaway food interventions

The World Health Organization (WHO) has identified that energy-dense and nutrient-poor foods, high in fat, sugar and salt, are contributing to an excess risk of chronic disease (World Health Organization, 2003). Recommendations and interventions have been implemented across the globe to challenge the rise in diet related non-communicable diseases (World Health Organization, 2010). In Australia, the methods used to impede takeaway food consumption have included a ban of fast food advertisements between 6am and 9pm, a ban on takeaway outlets opening within 400 meters of schools or leisure centres and taxes on high-fat fast foods and SSB (Obesity Australia, 2013). Alternative interventions have been implemented in the US; menu labelling of calories became law in 2010 as part of the Affordable Care Act (U.S. Food and Drug Association, 2016) for retail food chains with 20 or more establishments nationally (Cleveland et al., 2018). In New York, consumer awareness of the calorie information was assessed pre and post intervention and indicated that menu labelling on fast food generated a 2-fold increase in the percentage of customers making calorie-informed choices (Dumanovsky et al., 2010). Nevertheless, the US Food and Drug Administration extended the compliance date for implementing menu labelling of calories repeatedly during the previous 8 years and most recently to May 7th, 2018, due to non-compliance in some states; suggesting difficulty with implementing the intervention. However, a recent evaluation of calorie labelling at food chains revealed a high rate of compliance in advance of the May 2018 deadline showing feasibility of complying (Cleveland et al., 2018). Courtney et al. (2018) stated that calorie information may alter brain responses to food cues by simultaneously reducing reward system activation and increasing control system activation. Therefore, this type of intervention might facilitate healthier food choices in some individuals.

It is estimated that 25% kcal intake in the UK diet comes from out-of-home foods (Goffe et al., 2017) potentially contributing to outcomes such as weight gain, insulin resistance, and high blood pressure. Treating overweight and obesity-related ill-health costs the NHS in England £6.1 billion each year (Public Health England, 2017a), making it an area of high public health significance. The recent Global Burden of Disease study (Steel et al., 2018) showed that local authorities in the northern cities of Liverpool and Manchester had higher attributable risk and years life lost compared with authorities with similar deprivation in London and Birmingham. Reinforcing the need to respond to these distinct problems in northern cities through focused action (Whitehead, 2014; Minton and McCartney, 2018). In the UK, local government initiatives have aimed to tackle the impacts of takeaway food in communities via a multi-targeted approach. Such initiatives include working with the takeaway food industry to reformulate foods; reducing the amount of fast food consumed by school children; and addressing the proliferation of hot food takeaway outlets through planning regulations (Public Health England, 2017b; Public Health England, 2018a). In 2014, Newcastle City Council refused planning permission for a new drive-through McDonald's from opening near a local school in Kenton (BBC News, 2014). The area which had higher rates of obesity than the England average introduced fast food exclusion zones causing the fast food chain to withdraw plans (BBC News, 2016). It is now considered that there is enough evidence, despite some being conflicting, regarding the relationship between the built food environment and obesity for the development of specific planning policy

(Townshend and Lake, 2017). In order for strategies to be effective, it is considered vital that policy, practice, civic society and industry work collaboratively (Lake, 2018). Recent UK statistics have shown a 34% increase in the number of fast food outlets since 2010 (Office for National Statistics, 2018); providing further rationale to focus on local authority planning and health.

In April 2018, the UK government introduced the Soft Drinks Industry Levy, commonly referred to as the ‘Sugar Tax’, as a means of tackling childhood obesity through sugar reduction (Public Health England, 2018f). In addition, PHE published ‘Strategies for encouraging healthier out-of-home food provision’ (Public Health England, 2017b) and ‘the calorie reduction programme’ (Public Health England, 2018a) to encourage local intervention, including targets to reduce calories in takeaways through reformulation and portion size control, in a new obesity drive to cut calorie consumption by 20% by 2024. Previous research at LJMU investigated recipe reformulations to reduce sodium (Na) and fat content of takeaway foods and found that it was possible to reformulate a takeaway dish without decreasing consumer acceptability (Jaworowska et al., 2011). Even so, staff at fast food outlets engaging in "upselling" are trained to persuade customers to buy additional or larger portions (Royal Society for Public Health, 2018).

Nutrition information is regarded as essential when making healthy food choices and despite evidence showing the unfavourable nutritional content of takeaway foods (Jaworowska et al., 2014) this information has not been disseminated to consumers. Menu labelling is considered as a potential environmental strategy for addressing the obesity pandemic (Kerins et al., 2018). That said, there are currently public health strategies in place that provide advice, guidance, and encouragement on healthy eating (Adams et al., 2016) suggesting that consumer awareness of nutritional information may not be the issue. In the UK, the majority of independent takeaway food outlets do not provide kcal information to consumers and if fast food chains provide calorie information it is typically online or in inconspicuous locations. In response to the predominant absence of nutrient content of takeaway foods Public Health England (2017a) has urged takeaway and fast food outlets to introduce nutritional labelling. From September to December 2018, PHE opened up consultation seeking views on its plans to implement mandatory calorie labelling in out-of-home food outlets (Public Health England, 2018d). Research at LJMU investigating nutritional labelling of takeaway foods found that most consumers wanted information regarding nutrient content (Blackham et al., 2018b). Needless to say, participants may not make informed healthier choices and such strategies are unlikely to reduce inequalities in diet and obesity (Adams et al., 2016). An Australian study on energy and traffic light labelling of fast food found no impact on parent and child fast food selection (Dodds et al., 2013). Furthermore, the limitations of labelling takeaway meals, that may vary nutritionally on a daily basis and between establishments, makes implementing such an intervention extremely difficult (Blackham et al., 2018a). Therefore, novel alternative forms of nutritional labelling such as using physical activity labelling (Davies et al., 2015) require further investigation and development.

In order to create effective public health interventions in relation to takeaway food and obesity, it is necessary to have a strong foundation of primary evidence to support ideas. Dietary surveys such as the NDNS have previously included simple frequency questions on takeaway foods such as “On average, how often do you/does your child eat take-away meals (pizza, fish and chips, Indian, Chinese, burgers, kebab etc.) at home?" (Ziauddeen et al., 2017). As with all self-reported methods, dietary surveys are prone to limitations including recall bias, where an individual selectively recalls food items (Naska et al., 2017), over and under reporting and difficulty when estimating portion sizes. Dietary pattern analysis offers a more holistic approach to assess the dietary intake than nutrient analysis in more traditional studies. This method may counteract the inaccuracies and bias associated with dietary surveys. Moreover, the utilisation of a variety of methods to measure dietary exposure (self-report food frequency questionnaire (FFQ), interviewer led 24 hour 'Multiple Pass' dietary recalls (24 h MPR) and metabolite fingerprinting) to explain the current consumption in the target population may produce more valid results.

2.4 Food environment

Diet is a major environmental exposure and risk factor of a wide range of non-communicable diseases (Bingham, 2002; Shim et al., 2014). The food environment (or "foodscape"), extensively studied over the last 20 years, defines a major increase in out-of-home food establishments that is concordant with the proliferation of obesity (Burgoine et al., 2014). A review by Albuquerque et al. (2015) acknowledged the importance of genetic factors in the aetiology of obesity and inferred that natural selection has assisted the spread of genes that increase the risk for an obese phenotype. However, cumulatively all genomic markers along with their presumptive genes have only been shown to have a small effect on BMI (less than 5% of the total heritability) (Speliotes et al., 2010) and risk of obesity (Tan et al., 2014), further suggesting that obesity is more likely to be contextual (environmental influences that cause its inhabitants to become obese).

Environments that encourage the consumption of food and/or discourage physical activity have been labelled “obesogenic” (Reidpath et al., 2002) (Appendix 9.2). In Norfolk, UK, the number of takeaway outlets was reported to have grown by 45% between 1990 and 2008, a trend which has been reflected across the rest of the UK (Maguire et al., 2015). This abundance of unhealthy and energy-dense food in the environment, noted by Feng et al. (2010), has been shown to disrupt an individual’s ability to make healthy food choices (Maguire et al., 2015). A number of US studies have demonstrated that neighbourhood exposure to fast food outlets increased consumption near the home in addition to contributing to a poor diet (Moore et al., 2009; Athens et al., 2016) (Appendix 9.2). A prospective one-year study by Li et al. (2009) found that neighbourhoods with a high density of fast food outlets promoted an increase in weight and waist circumference in those who visited frequently. Regardless of the link between neighbourhood availability of takeaway food and higher BMI/greater odds of obesity (Burgoine et al., 2009; Burgoine et al., 2014), other studies have challenged this. For example, Turrell and Giskes (2008) showed no relationship between the purchasing of takeaway food, road distance to the closest takeaway outlets and the number of takeaway outlets in the local food environment of Brisbane, Australia. They found that dietary inequalities between socio-economic groups appeared to have a stronger influence on the purchasing of takeaway food (Turrell and Giskes, 2008). This suggests that the food environment may be more complex, with economic and socio-cultural factors potentially influencing food consumption and food-related behaviours (Giskes et al., 2011).

The many studies referring to “obesogenic environments” make simple correlations between environment and obesity and do not explore the sociological and behavioural determinants of food consumption. For example, a large study by Pieroni and Salmasi (2014) stated that there was a clear correlation, but no causal relationship, with the higher availability of fast food outlets and increased BMIs (Appendix 9.2). In a more recent study, Polsky et al. (2016) reported an increase in obesity figures among adults living in close proximity to a number of fast food outlets; suggesting that a food environment with a high density of fast food outlets is most likely to impact on weight status (Appendix 9.2). Overall, the literature suggests that the food environment is an important factor to consider when contemplating the reasons for takeaway food consumption and is a potential target for change. However, age group, socio-economic status and culture are other factors considered as important influences and without conducting a randomised control trial, it is impossible to differentiate the cause and effect within a cross-sectional design study. Likewise, the food environment is regarded as merely one factor in the causes of obesity, which are complex and multifaceted (Butland et al., 2007) (Fig. 2.1).

2.5 INEQUALITIES, TAKEAWAY FOOD AND HEALTH

An unequal distribution of health – geographically, ethnically and socially, has detrimental effects on those of low socio-economic status (Wilkinson and Pickett, 2010). Studies in the UK investigating neighbourhood deprivation and access to fast food outlets have found an association with increased levels of obesity (Cummins et al., 2005; Macdonald et al., 2007; Maguire et al., 2015). A current

UK report showed a strong link between deprivation and density of fast food outlets, with deprived areas such as Blackpool and parts of Manchester and Liverpool having five times more fast food outlets than the least deprived areas (Fig. 2.2) (Public Health England, 2018b). The disproportionate concentration of fast food outlets in deprived areas is particularly prevalent in the north of England, where levels of deprivation are typically higher than in the south (Buchan et al., 2017). However, similar trends are present across the UK, in deprived areas where the demand for inexpensive energy-dense food is often higher (Royal Society for Public Health, 2018; Wise, 2018). These findings corroborate those in Australia (Reidpath et al., 2002), New Zealand (Pearce et al., 2007; Utter et al., 2011), Germany (Schneider and Gruber, 2012), Canada (Smoyer-Tomic et al., 2008), and the US (Zenk and Powell, 2008), who observed that those living in the poorest areas had a higher exposure to fast food outlets than those in less deprived areas (Appendix 9.2). In contrast, high socio-economic status and urban residence was associated with consumption of energy-dense foods in adolescents in China (Shi et al., 2005); suggesting accelerated nutrition transition towards a western diet within communities experiencing economic growth. Yet, in West Africa (Zeba et al., 2012), Bangladesh (Shafique et al., 2007), and Indonesia (Hanandita and Tampubolon, 2015), income inequality and economic development has been shown to increase the odds of a double burden of malnutrition; the coexistence of both under- and overweight.

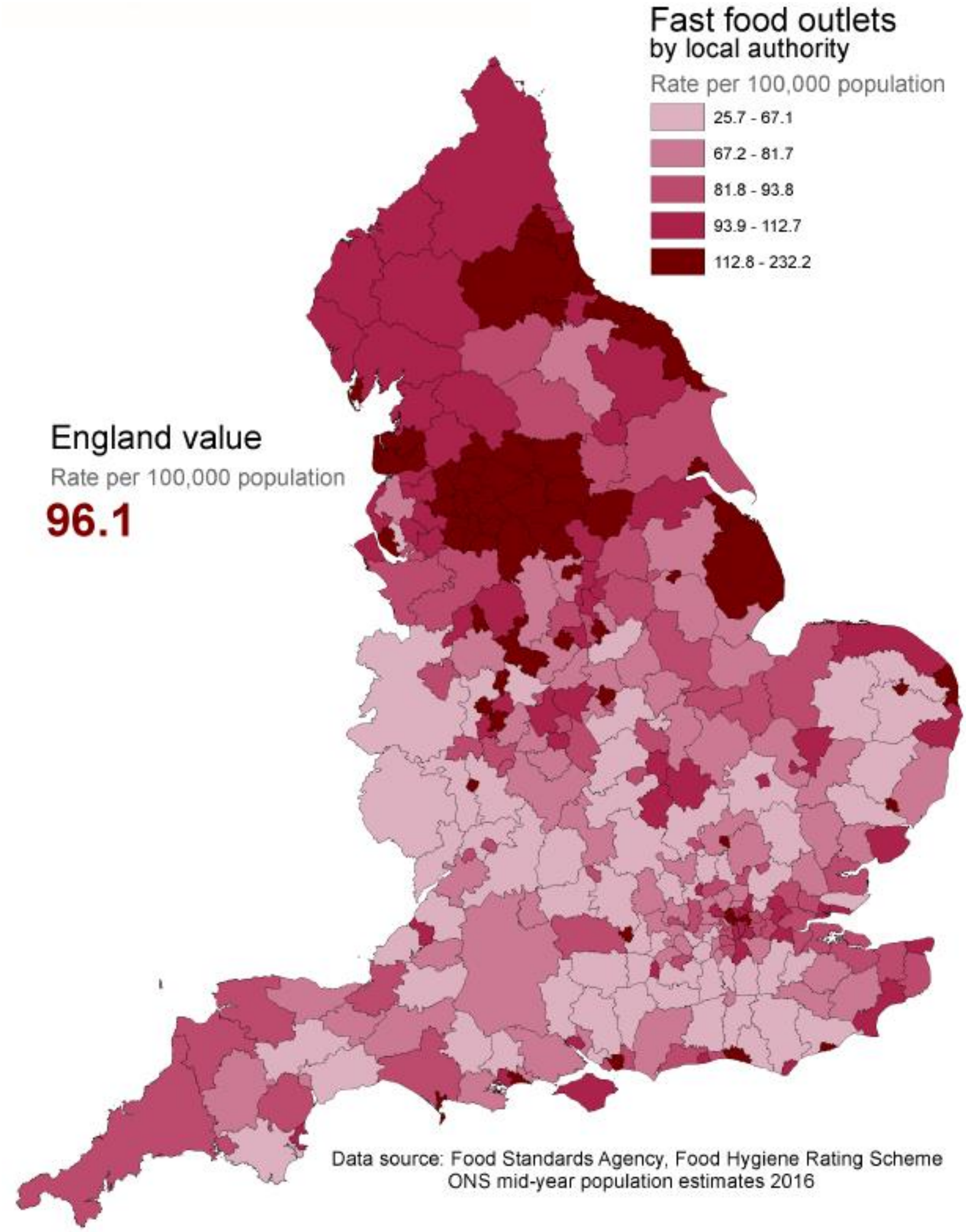


Figure 2-2 Density of fast food outlets in England by local authority (Public Health England, 2018e)

In the US, rates of obesity and poor health were most prevalent in the least educated and poverty-stricken population groups (Drewnowski and Specter, 2004; Braveman et al., 2010). Nevertheless, data from the National Longitudinal

Survey of Youth in the US showed no relationship between fast food consumption and socio-economic status (Zagorsky and Smith, 2017). Therefore, suggesting that poor health outcomes in lower socio-economic groups are not solely due to fast food intake but could be a marker of poor habitual diets and other lifestyle factors. In Scotland, UK, consumption of takeaway food was significantly higher in the most deprived quintile (Barton et al., 2015). A recent UK study (Goffe et al., 2017) using NDNS data also found that the mean energy intake of children from less affluent households was more positively associated with takeaway meals than those from more affluent households. Therefore, children living in more deprived areas may not only be exposed to more takeaway outlets but also be more susceptible to the effects of consuming takeaway food (Goffe et al., 2017). Research from Australia investigating the frequency and types of takeaway foods consumed by different socio-economic groups found that individuals from disadvantaged groups were consistently consuming less healthy takeaways, classified based on the Australian Guide to Healthy Eating, than those from advantaged groups (Miura et al., 2009; Miura et al., 2012) (Appendix 9.2). Lake et al. (2007) explored perceptions and practice of healthy eating and revealed that individuals from a higher socio-economic group were more likely to agree with the statement ‘my eating patterns are healthy’. Despite some conflicting findings between the effects of socio-economic position on the food environment and takeaway consumption, the greater part of the literature suggests that those from lower socio-economic groups would be more susceptible to inequalities in diet and as a result increased rates of obesity and chronic disease.

Other studies on the socio-economic disparities in the food environment have concentrated on the notion of food security and insecurity. Food security was defined by the 1996 World Food Summit as “a situation that exists when all people, at all times, have physical, social and economic access to sufficient, safe and nutritious food that meets their dietary needs and food preferences for an active and healthy life” (Food and Agriculture Organization, 1996). Food insecurity, on the other hand, refers to the limited access to affordable, quality, and nutritious food, but also restrictions on the facilities to store, cook and consume those foods (Borch and Kjaernes, 2016). A mail survey on adults from disadvantaged suburbs of Brisbane city, Australia, showed that approximately one in four households were food insecure based on results from an eighteen-item food security screening questionnaire (Ramsey et al., 2012). The economic and physical access constraints to nutritious food in deprived areas have contributed to what are sometimes defined as ‘food deserts’ (Whelan et al., 2002; Wrigley, 2002). A particularly interesting finding from the Brisbane study was that food-insecure households were two-and-a half times more likely to report more frequent hamburger consumption compared with those who were not food insecure (Ramsey et al., 2012). These findings support the notion that food insecurity may encourage the purchasing of takeaway food; especially in deprived areas (Hendrickson et al., 2006). Thus, targeting areas of high deprivation and ensuring food security may be a strategy for facilitating healthy eating (Trapp et al., 2015).

Less healthy takeaway food choice has been shown to be associated with a poorer level of education (Miura et al., 2009; Miura et al., 2012). However, research suggests that poor health literacy is a stronger predictor of health than variables such as age, ethnicity, income, employment status and education level (Parker, 1999). Health literacy refers to an individual’s knowledge and skills in matters of health and illness (Nutbeam, 2008). Poor health literacy is recognised as a cause of health inequalities in both rich and poor countries (Marmot and Commission on Social Determinants of Health, 2007). Boulos (2005) stated that most written resources containing health information were deemed too advanced for the general UK population, with an average reading age of nine. Thus, it was found that limited health literacy was related to unhealthy lifestyle behaviours such as poor diet (Public Health England, 2015). An emerging concept is food literacy that encompasses individual food skills, community food security and health literacy (Cullen et al., 2015). Carbone and Zoellner (2012) specified that literacy was a determinant of dietary patterns and that increased food literacy was positively associated with healthier eating practices. For example, a study on young adults showed that those with low levels of health literacy used food labels significantly less (Cha et al., 2014), suggesting that their food choices were less informed by nutritional information. Therefore, it would appear crucial to consider literacy levels when conducting any takeaway food intervention or research. Some nutrition studies include validated health literacy assessments to understand participant knowledge, their interpretation and behaviour towards aspects, such as Nutrition Facts label (Zoellner et al., 2012). Nevertheless, nutrition labelling at independent takeaway outlets may not be plausible due to significant variation in the nutrient content of identical meals and inconsistent portion sizes (Jaworowska et al., 2014). In addition, alternate chefs with different cooking styles and the absence of formal recipes introduces further limitations of food labelling in takeaway outlets (unpublished data). Moreover, even if the population had increased health or food literacy, conflicting food messages from a myriad of sources makes it challenging for society to make healthy choices.

Economic disadvantage in the food environment appears to be a strong determinant of access to takeaway foods and consequent intake. For individuals on a very low income it appears to be considerably more difficult to make healthy food choices than for their more affluent counterparts. Therefore, there is a warrant for further research to understand socio-economic differences between types and frequencies of takeaway food intake.

2.6 Socio-demographic determinants of takeaway food consumption

Previous research has categorised takeaway food consumption according to gender. A study on Australian adults, found men consumed takeaway foods more frequently than women (Smith et al., 2009) (Appendix 9.3). This could be explained in part by findings showing that gender was the strongest determinant of time spent cooking at home (as mentioned previously), with women spending more time cooking than men (Adams and White, 2015). Mercille et al. (2016) found that older men's dietary patterns were associated with cues for fast food outlets whereby they were heavily influenced by external exposure to fast food when making food choices (Appendix 9.3). Other studies observed that men received a higher proportion of their energy intake from foods prepared and consumed out of the home (Orfanos et al., 2009; Vandevijvere et al., 2009). However, a UK study found that the only gender difference was seen in children; boys consumed more takeaway meals at home than girls (Adams et al., 2015) (Appendix 9.3). This suggested that alternative patterns of out-of-home eating existed in the UK or had altered in recent years (Adams et al., 2015). Yet, contrasting data from 2014 showed that men were more likely than women to report eating fast food (27% compared with 16%) and takeaways (32% compared with 22%) (Food Standards Agency, 2014). Other findings have suggested that women and older adults are more vulnerable to higher BMI, in addition to statistically lower serum HDL-cholesterol (increasing CVD risk) and concentrations of micronutrients, with increased consumption of out-of-home foods (Kant et al., 2015). That said, results showed no statistical support that consumption of out-of-home foods altered profiles of other biomarkers linked to greater metabolic risk (Kant et al., 2015). The Australian study by Smith et al. (2012) exposed an increased risk of cardiometabolic disease in young women who consumed takeaway foods twice a week or more. Therefore, despite some studies suggesting that younger males consume takeaways more frequently, the health implications may not be as severe as those seen in females or those in older adults. The increased risk to females and older adults may be attributable to lower amounts of physical activity and/or a lower basal metabolic rate than younger males. However, this is the current authors' speculation and thus requires further investigation.

In the UK, the consumption of meals out-of-home and takeaway meals at home was particularly widespread among the younger age groups and was shown to peak in those aged between 19 and 29 years (Adams et al., 2015). Previous studies reported comparable findings in other European countries (Orfanos et al., 2009; van der Horst et al., 2011), the US (Powell and Nguyen, 2013; Athens et al., 2016), and New Zealand (Smith et al., 2014). Likewise in Australia, consumption of takeaway and fast food was shown to increase from adolescence to young adulthood (Smith et al., 2009) and a relatively high consumption of fast food occurred between the ages of 18 and 45 years (Mohr et al., 2007; Dunn et al., 2008; Dunn et al., 2011) (Appendix 9.3). In China, people aged 15–39 years spend the greatest on out-of-home food and consume the most (Liu et al., 2015). Many diet-related health issues stem from adolescence, a time when young people require an increase in nutrients (Neumark-Sztainer et al., 2004; Luszczynska et al., 2013) but often make unhealthy choices (Dwyer, 2001; Slining et al., 2013). Fast food, culturally important to adolescents, is one of the limited types of food that is affordable amongst that group. Furthermore, the types of food consumed by young people are an important symbol of social and cultural belonging (Bugge, 2011). A Swiss study that examined the importance of balanced food choices suggested that lack of cooking skills may play a part in driving younger age groups to consume more takeaway foods (Hartmann et al., 2013) (Appendix 9.3). Statistics have shown a downward trend in consumption of both meals out and takeaway meals at home in older adults (Adams et al., 2015). Older age groups may have less disposable income (Banks et al., 2010) and may find takeaway foods unfamiliar, with a lack of exposure in younger years when eating habits develop (Kant and Graubard, 2004).

In Los Angeles, US, areas with a high population of immigrants lacking acculturation were associated with healthier dietary behaviour (Zhang et al., 2015). Nonetheless, once immigrants become acculturated unhealthy dietary behaviours are adopted (Gordon-Larsen et al., 2003; Unger et al., 2004; Pérez-Escamilla and Putnik, 2007; Batis et al., 2011; Kaiser et al., 2015). With respect to ethnic groups Block et al. (2004) showed that fast food outlets in New Orleans, US, were geographically associated with predominately black and low-income neighbourhoods after controlling for environmental confounders (commercial activity, presence of highways, and median home values). Correspondingly, a study in Texas, US, found that non-whites exhibited higher obesity rates, increased availability of fast food establishments in their local environment and higher consumption of fast food meals than their white counterparts (Dunn et al., 2012). A study on the variations in fast food consumption in India showed that Indians preferred fast food from global chains compared to Indian fast food because they said that global brands were of better quality (Srivastava, 2015) (Appendix 9.3). Likewise, minority ethnic groups of females living in the UK have incorporated the less healthy aspects of the Western diet including fast foods (such as fried fish, pizza, fries and fatty snack foods) into their diet when time was limited (Lawrence et al., 2007). However, a study in Bradford, UK, found a negative association between BMI and fast food outlet density in a South Asian Group of women (Fraser et al., 2012) (Appendix 9.3); which would indicate a lack of acculturation. Therefore, showing evidence on ethnicity remains equivocal; indeed El-Sayed et al. (2011) found that there was a lack of consensus regarding the aetiology and relative risk of obesity among large ethnic minority groups when compared to Caucasians in the UK. Furthermore, Fraser et al. (2012) argued that there was little research conducted outside of the US to explain whether ethnicity was related to access to and consumption of fast food. Additionally, ethnic minorities are disproportionately represented in low-income areas, thus socio-economic position is a confounding variable. Therefore, the limitations related to current research on ethnicity as a determinant of takeaway food consumption warrants further investigation.

2.7 DIETARY ASSESSMENT AND DIETARY PATTERNS

Dietary assessment has typically used nutrient analysis to analyse population intake. However, research has shown a diversity in nutritional composition, both between and within types of takeaway food (Jaworowska et al., 2014) suggesting that nutrient analysis may not be suitable for assessment of takeaway food consumption. Simple frequency of takeaway food consumption (Adams et al., 2015) does not account for the impacts of the remainder of the diet. Moreover, understanding the subtle differences in takeaway food intake may be more predictive of health outcomes than frequency of consumption. For example, a habitual diet containing energy-dense nutrient-poor foods and high-calorie fast foods could impose increased health risk than a habitual diet containing nutritious foods and lower-calorie takeaway options. Thus, there is a need for a better approach to analyse takeaway food consumption and habitual diet.

In recent years, research into dietary intake has shifted from analyses of single food or nutrient consumption to a more complex assessment of the whole diet to produce dietary patterns (Fung et al., 2001; Cespedes and Hu, 2015). The significance of dietary patterns is due to the consequent synergistic effects of food and nutrient combinations (as opposed to individual foods or nutrients) which may be more predictive of disease risk and aid prevention of obesity and non-communicable disease (Hu, 2002; Granic et al., 2015). Moreover, despite nutrient analysis being common practice it is often misleading to assume that single nutrients or foods can be observed in isolation in relation to disease risk (Jacques and Tucker, 2001). Instead, dietary patterns embrace the comprehensive relationship between nutrition and disease and addresses issues of collinearity of nutrient intake and interactions (Gibbons et al., 2017).

The most common method to derive dietary patterns is a hypothesis-driven approach known as *a priori* that uses diet indices or scores to assess adherence to a specific diet (i.e. Mediterranean Diet Score or Dietary Approaches to Stop Hypertension (DASH)), to a healthy eating index or to dietary guidelines (Granic et al., 2015) (Appendix 9.4). This technique relies on the accuracy of the definition of a healthy diet, which may not be representative of different populations that are forever evolving and changing. The *a priori* scores all tend to promote the intake of fruits, vegetables and whole grains (Hodge and Bassett, 2016). The exploratory (data-driven) approach to dietary pattern analysis known as *a posterori* does not rely on the current field of nutrition knowledge and may provide more accurate descriptions of the habitual diets of specific population groups (Granic et al., 2015). Instead, this approach relies on statistical analysis such as factor or cluster analysis to produce population specific dietary patterns.

Previous *a posterori* research has predominately produced habitual dietary patterns including a Western (unhealthy) dietary pattern and a Prudent (healthy) dietary pattern to describe population diets and associated health outcomes (Fung et al., 2004; Bertuccio et al., 2013; Stricker et al., 2013). The Mediterranean dietary pattern (components including vegetables, legumes, fruits, nuts, whole grains, fish, high monounsaturated fat and low saturated fat rich food, alcohol, and meat) has been heavily investigated for its acclaimed beneficial role on health and longevity (Couto et al., 2011; Kastorini et al., 2011; Rees et al., 2019; Estruch et al., 2013; Lopez-Garcia et al., 2014). Findings from a multicenter randomised trial in Spain found that a Mediterranean diet supplemented with extra-virgin olive oil or nuts reduced the incidence of major cardiovascular events in individuals with high CVD risk (Estruch et al., 2013). In addition, a large prospective cohort study on a US population provided strong evidence that the Mediterranean dietary pattern was associated with decreased all-cause mortality including CVD and cancer (Mitrou et al., 2007). A cross-sectional study on Nepalese adults used principal component analysis (PCA), a type of factor analysis, to derive dietary patterns from FFQ data and to investigate associations with overweight and obesity (Shrestha et al., 2016). They produced four distinct dietary patterns including a fast food dietary pattern, mixed dietary pattern, refined grain-rice-alcohol dietary pattern and solid fats-dairy dietary pattern. The refined grain-rice-alcohol dietary pattern was significantly associated with overweight adults and the fast food dietary pattern was associated with obesity in adults (> 40 years) (Shrestha et al., 2016).

A study on Australian adults, assessed takeaway food consumption by asking consumers how often they usually ate twenty-two specific takeaway food items (Miura et al., 2012). The takeaway food items were classified as ‘healthy’ and ‘less healthy’ based on the Australian Guide to Healthy Eating and were used to examine socio-economic differences in the frequency and types of takeaway foods consumed (Miura et al., 2012). Whilst this study was able to establish links between socio-economically disadvantaged groups and unhealthier takeaway food choices, there was no inclusion of habitual diet to demonstrate what role takeaway foods had on their overall diet. For instance, some authors have speculated that regular takeaway food consumers are more likely to have an overall unhealthy dietary pattern. Poti et al. (2014) found a correlation between children who consumed fast food and a Western dietary pattern and suggested that the remainder of the diet could be more predictive of overweight/obesity and poor dietary outcomes than fast food consumption itself. This may be attributable to fast food consumption negatively effecting satiety, which could potentially increase consumption of other foods in the remainder of the diet, including more Western food. Nevertheless, this is the current authors' speculation and thus requires further investigation.

There has been no research conducted in the UK that has explored the dietary patterns of takeaway food consumers and what role this may bare on diet and health. Thus, research into this area could provide valuable insight into takeaway food consumers and highlight areas for further work.

2.8 METABOLITE FINGERPRINTS IN URINE

Accurate measurement of dietary intake is an essential component of much nutrition and health-related research (Fave et al., 2009). Dietary assessment techniques including FFQ, 24 h MPR and food diaries are common practice but have a number of limitations including recall bias, and under and over reporting that can cause unreliable estimates of food and nutrient intakes (Naska et al., 2017). Moreover, such techniques are not always appropriate for use on certain populations. Previous research has used urinary nitrogen, potassium, and sodium biomarkers as a validation technique for exposure to some foods (McKeown et al., 2001). Dietary biomarkers are found in a range of human biofluid samples including blood, urine or saliva, and can be used to indicate dietary exposure and to improve the accuracy of estimated consumption by self-reported dietary assessment methods (Woodside et al., 2017). Currently dietary exposure biomarkers are limited to a relatively small number of specific foods and food components and most are of uncertain validity (Lovegrove et al., 2004; Lee et al., 2006; Marklund et al., 2010).

The rapid development of metabolomics has been the result of advances in instruments, progress in machine learning data analysis using high performance computation, and increased access to metabolite annotation and structural databases (Scalbert et al., 2014). As a result, there has been recent identification of novel biomarkers for many foods including fruit, vegetables, beverages, meats and complex diets (Scalbert et al., 2014). A study on participants (n = 481) from the large EPIC cohort measuring metabolite fingerprints using high-resolution mass spectrometry in 24 h urine samples, and 24 h dietary recalls and FFQs, identified 6 novel biomarkers of polyphenol-rich foods (coffee, tea, red wine, citrus fruit, apples and pears, and chocolate products) (Edmands et al., 2015). The advantage of using non targeted approaches such as flow infusion-high resolution fingerprinting (FIE-HRMS) is that any potentially ‘explanatory’ (significant and biologically relevant in context) m/z signals may be linked directly to a specific metabolite by atomic mass information (Dunn et al., 2005; Beckmann et al., 2008; Enot et al., 2008). An epidemiological study involving 4,630 participants from China, Japan, the UK and USA using four in-depth 24 h MPR, two 24 h urine samples and PCA produced urinary metabolite excretion patterns that were significantly different for East Asian and western populations (Holmes et al., 2008). This highlighted that metabolomics can explain the subtle differences between individuals or populations exposed to different diets, lifestyles and environments.

Several reports have described the analysis, in human biofluids, of specific metabolites known to be derived from foods of high public health significance such as wholegrains or citrus fruits (Lloyd et al., 2011; Beckmann et al., 2013), but very few validated biomarkers for takeaway and fast foods (Beckmann et al., 2016). A recent review on dietary biomarkers in nutritional epidemiology identified three major applications for the technology; determination of food intake using biomarker levels from feeding studies, dietary patterns of individuals using urinary metabolic fingerprints and metabolome-wide-association studies (Brennan and Hu, 2018). However, the authors concluded that further metabolomics research is warranted for the use of biomarkers in dietary assessment (Brennan and Hu, 2018). Further development of this biomarker technology has the potential to enhance nutritional research by improving the accuracy of dietary assessment, testing compliance to dietary guidelines and investigating associations with disease outcomes (Hedrick et al., 2012).

2.9 RATIONALE

The current cross-sectional observational study aims to investigate takeaway food consumption and wider dietary patterns and the factors influencing their consumption. Additionally, to identify novel fingerprint patterns, and metabolite biomarkers, which are associated with takeaway/fast food diets/unhealthy eating. The data from this cross-sectional study will add to a coherent body of evidence on takeaway food from a 10 year period at LJMU, by complimenting the existing evidence on nutritional composition and behaviours relating to takeaway foods. To date, the research at LJMU has focussed on the nutritional composition (Jaworowska et al., 2014), reformulation (Jaworowska et al., 2011), nutritional labelling (Blackham et al., 2018b) and physical activity equivalents (Davies et al.,

2015) of takeaway foods. Thus, the primary focus has been on the takeaway foods themselves. The current study will address the takeaway food consumers.

This study is of particular importance as it incorporates a complete range of demographics (self-reported socio-demographics, anthropometrics and lifestyle characteristics) compared to other studies. Moreover, it includes several layers of detailed dietary assessment (FFQ, 3 x 24 h MPR and metabolite fingerprinting) conducted in the relatively un-researched area of Merseyside in North West England. This geographical area has a high density of takeaway outlets (UKCRC Centre for Diet and Activity Research (CEDAR), 2017b) where takeaway food has been shown to have an undesirable nutritional content (Jaworowska et al., 2012; Jaworowska et al., 2014). Furthermore, the methodology for this proposed study includes an innovative dietary pattern analysis using PCA on habitual and takeaway foods. Dietary patterns are more suitable for exploring population intake at a multidimensional (i.e., it is a complex, multi-layered exposure and behaviour) and dynamic (i.e., it varies over time and the life course) level, compared to more traditional methods that analyse single nutrients (Reedy et al., 2018) (Appendix 9.4). Thus, PCA is favourable to account for the subtle differences in wider dietary patterns, without focusing on the nutrient content. PCA is an existing advanced statistical technique that has been used to progress nutritional research. This project will use PCA in an original way due to the exploratory nature of the data. In addition, this project comprises an innovative metabolite fingerprinting technology on urine samples from takeaway food consumers.

This doctoral thesis aimed to assess the dietary patterns of takeaway food consumers and links to socio-demographic and lifestyle factors by investigating the dietary intake of a sample of the Merseyside population and performing PCA on collected data. This study also aimed to validate dietary assessment techniques by using two forms of self-reported dietary assessment and one biomarker technique. Finally, this study aimed to classify metabolite fingerprints in urine indicative of takeaway foods/unhealthy foods and to add to a model for objective classification of dietary exposure based on urinary metabolite fingerprint data.

Chapter 3
General Methods

3 Chapter 3: General Methods

3.1 Chapter outline

This PhD project combines a broad spectrum of methods including multiple dietary assessment techniques, dietary pattern analysis and metabolomics to form a coherent scientific project. Other noteworthy techniques have included use of social media for recruitment and development of a bespoke urine sampling kit for spot urine sample collection. A sample of adults (aged 18–64 years), recruited from Liverpool John Moores University (LJMU), participated in the pilot stage (Study 1) of the research. Participants recruited from Merseyside participated in the main stage of the research (Study 2) and a sub-sample of self-selected individuals participated in the final stage (Study 3). Recruitment and data collection initiated in January 2016 and ended in October 2017. This chapter provides an overview of the study design and the individual study chapters (4, 5 and 6) provide further detail. Further information regarding the methods used are discussed below.

3.2 Overall study design

To meet the aims and objectives in Chapter 1 (Page 23-24), the overarching study design included the collection of multiple layers of retrospective dietary assessment and metabolomics data to provide a holistic overview of the population in terms of takeaway food consumption, dietary patterns and their associated determinants. The pilot (Study 1) gathered feedback on study design, tested the methods for Study 2 and acted as a recruitment feasibility study. The pilot included two measures (food frequency questionnaire (FFQ) and three 24 hour ‘Multiple Pass’ dietary recalls (3 x 24 h MPR)) to assess habitual and takeaway food consumption, to record detailed dietary intake and to validate methods. The cross-sectional study (Study 2) comprised a comprehensive FFQ to assess habitual and takeaway food consumption and an extra section to record socio-demographic background of a Merseyside population. The final exploratory study (Study 3) recorded detailed dietary intake and explored metabolite fingerprints from urine samples to validate food exposure, with the potential to identify markers of specific foods on a sub-sample of participants from Study 2.

This project was designed to collect dietary data from both short-term (3 x 24 h MPR) and long-term (FFQ) methods to allow for maximizing the strengths of each dietary assessment technique (Subar et al., 2015). It has been common practice to compare a FFQ against another dietary assessment method in previous validation studies (Cade et al., 2007). To validate the findings from the FFQ the use of 3 x 24 h MPR was used to analyse food consumption. If the findings were concordant with those from the FFQ, it would be concluded that the 3 x 24 h MPR validated the FFQ. A third objective measure (metabolomics) for biomarker discovery in urine assessed the validity of dietary intake and dietary patterns. The metabolomics research was integrated into the project from a collaboration with Aberystwyth University (AU) and involved multi-site organisation and production of a materials transfer agreement (MTA) (Appendix 9.26). An internal collaborator (Professor in Pharmacy and Biomolecular Sciences) assisted the temporary storage of urine samples at LJMU and the external collaborators (headed by the Institute Director of Enabling Technologies, AU) organised sample transfers, preparation of samples and analysis. A full method flowchart is outlined below for Study 2 and 3 (Fig. 3.1).

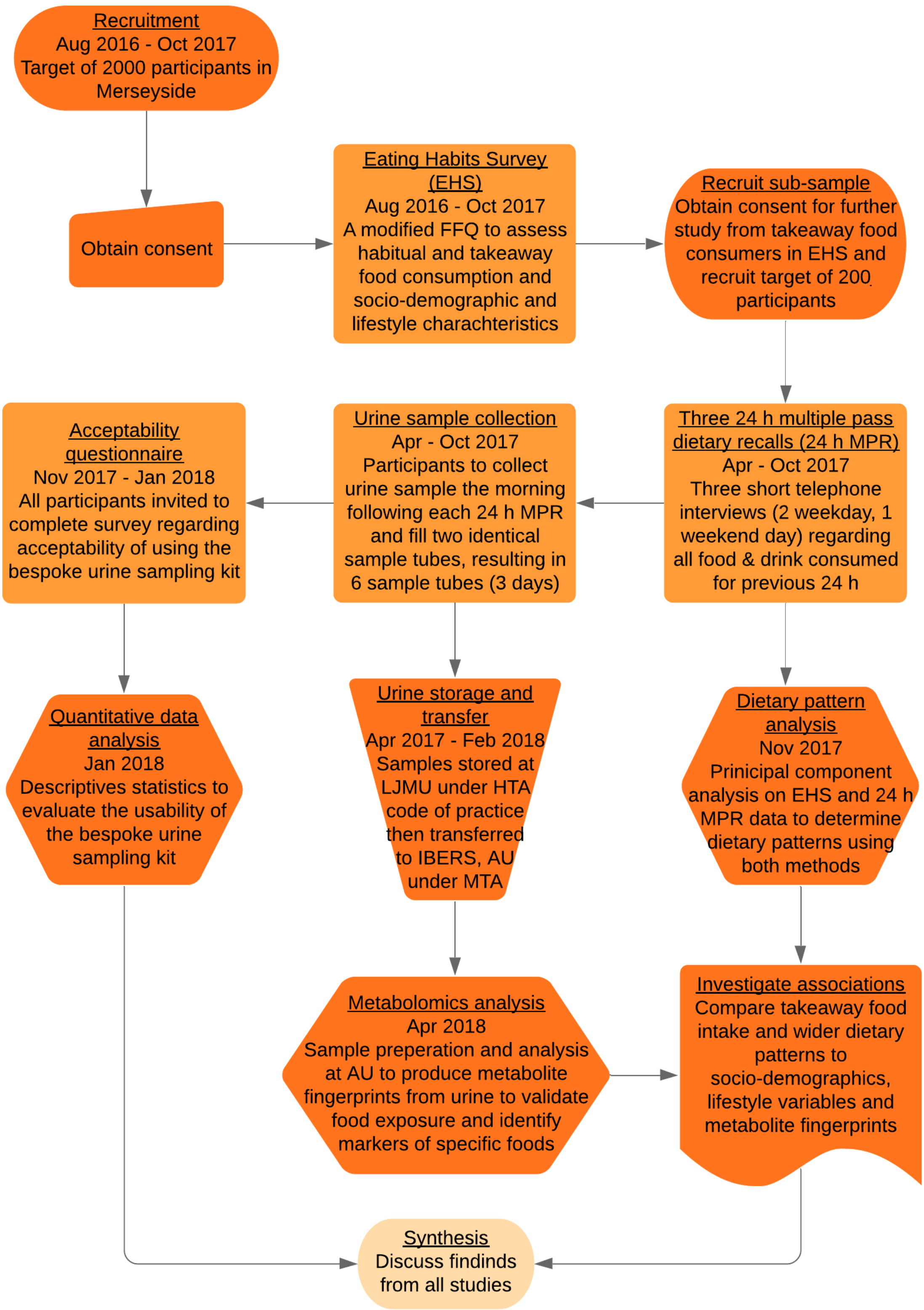


Figure 3-1 Full method flowchart for Study 2 and 3

3.3 ETHICS STATEMENT AND CONSENT

This study was conducted according to the guidelines laid down in the Declaration of Helsinki and the research ethics committees at LJMU approved all procedures involving human participants on the 11th January 2016 (16/EHC/002) (Appendix 9.5). The committee approved a major amendment on 10th March 2016 (Appendix 9.6) for the inclusion of urine sampling and metabolomics as part of the collaboration with researchers from the Institute of Biological, Environmental & Rural Sciences (IBERS) at AU. This ethics amendment involved receiving Human Tissue Authority (HTA) training and approval for the collection and temporary storage of urine samples at LJMU. In addition, this amendment included an update of study materials and recruitment methods (increase recruitment area from Liverpool to Liverpool and surrounding areas and use of social media for recruitment). Approval for further major amendments for changes made to recruitment methods (increase recruitment area from Liverpool and surrounding areas to Merseyside) occurred on 4th October 2016 (Appendix 9.7). Participant information was added to the online survey and a new flyer/poster was created on 10th November 2016 (Appendix 9.8). Changes to materials for Study 3 (including participant information, consent forms and photographic instructions) occurred on 7th February 2017 (Appendix 9.9) and the addition of an acceptability questionnaire for the bespoke urine sampling kit on 9th November 2017 (Appendix 9.10). Each participant gave written informed consent for Study 1, implied consent via the online survey for Study 2, written informed consent for Study 3 and implied consent for the online survey for Study 3.

3.4 STUDY PARTICIPANTS

Participant recruitment for Study 1 took place at LJMU between January and April 2016 via e-mail and face-to-face invites. Participant recruitment for Study 2 was conducted from August 2016 to October 2017 and involved a multi-pronged approach (social media, posters/flyers, word of mouth, newspaper advert etc.) to attract as many individuals as possible. Eligible participants (takeaway food consumers that provided contact details) in Study 2 were invited to participate in Study 3 from April to October 2017. Merseyside was selected as the study area to continue on from previous takeaway food research (Jaworowska et al., 2014), due to the large number of takeaway outlets identified by CEDAR (2017b) and somewhat for convenience. Moreover, obesity rates, cardiometabolic risk and social inequalities were higher in Liverpool than the England average (Public Health England, 2018c; Steel et al., 2018). All participants were aged between 18 and 64 years (the working age) at the time of recruitment. Figure 3.2 outlines a flowchart of participant recruitment and retention throughout the project.

Three £200 high street vouchers were purchased as incentives for the research. A prize draw for the first £200 voucher was held for the initial 1000 participants in Study 2 to incentivise further individuals to participate. A random code generator determined the winner who was contacted by e-mail and given 7 days to claim their prize; the vouchers were then posted. Dr Barbara Walsh, the Director of the School of Sport Studies, Leisure and Nutrition at LJMU, presented the prize as an impartial person to the project. At the close of Study 2, a second prize draw for a £200 voucher was held for the remaining 782 participants. The final £200 voucher prize draw was held for the 154 participants in Study 3.

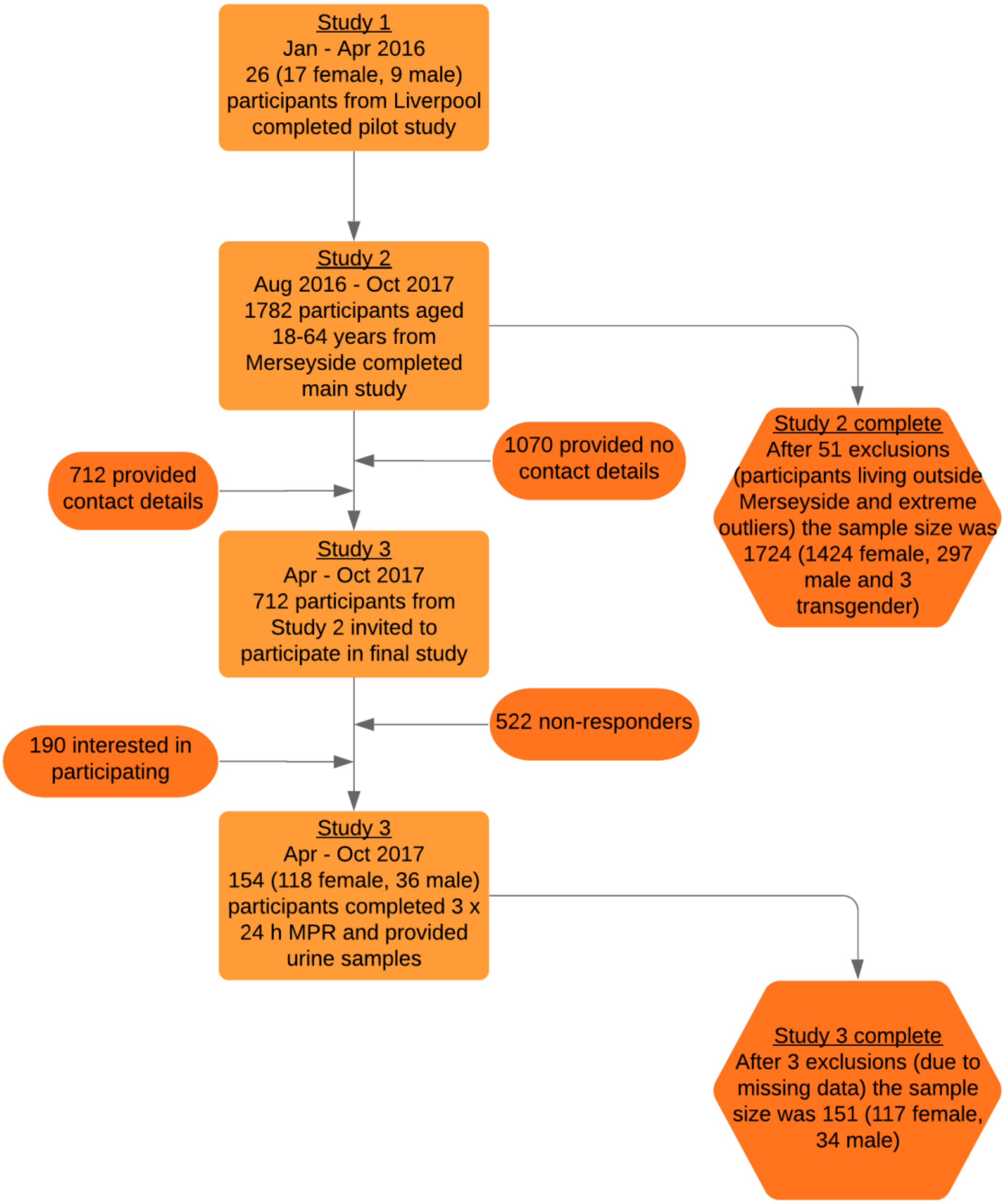


Figure 3-2 Flowchart of participants throughout the project

3.5 STUDY SAMPLE SIZE

A power calculation was not performed for the initial study as the research acted as a pilot and time was limited. The pilot study used convenience sampling because the purpose was to test methods and gather preliminary feedback to support the design of Study 2. A sample size of 20 participants was estimated as a realistic target for the pilot due to the time available for recruitment and data collection. For Study 2 a power calculation of 261 was completed for an accurate representation of takeaway food consumers in Merseyside (Table 5.1, Study 2, Chapter 5) (Naing et al., 2006; Charan and Biswas, 2013). However, previous dietary pattern studies included larger sample sizes of between 700 and 5,000 (Mullie et al., 2010; Appannah et al., 2015; Granic et al., 2015; Batis et al., 2016) (Appendix 9.4). Therefore, a target sample of 2000 participants was estimated for the cross-sectional Study 2. An additional factor that determined the target sample size for Study 2 was to have a large enough sample to recruit a sub-sample of 200 regular takeaway food consumers for participation in Study 3; the target set by researchers at AU.

3.6 STUDY DATA COLLECTION

3.6.1 FOOD FREQUENCY QUESTIONNAIRE (EATING HABITS SURVEY)

One of the main motives for conducting the pilot study was to develop appropriate dietary assessment tools to investigate takeaway food intake and produce dietary patterns. FFQs assess habitual diet by enquiring about the frequency with which food items or specific food groups are consumed over a reference period (i.e. one year). A number of studies have used FFQs to produce dietary patterns (van Dam et al., 2003; Zheng et al., 2016; Mertens et al., 2017) (Appendix 9.4). However, there had been limited cross-sectional studies in the UK regarding specific takeaway food intake therefore, a FFQ was designed for the purpose of this project.

3.6.2 24 HOUR 'MULTIPLE PASS' DIETARY RECALL (24 H MPR)

The 24 h MPR method (Thompson and Subar, 2008) captures individuals' diet by recording detailed food intake during the previous day (Granic et al., 2015). This method has been used extensively in national surveys in the UK and the US (Nelson et al., 2007; Moshfegh et al., 2008) and provides more accurate estimates of energy and nutrient intakes compared with the FFQ (Adamson et al., 2009); thus acting as an ideal validation technique. Due to the day-to-day variation in individual food intakes, it is recommended to use a number of days of intake data to determine usual food intakes and improve reliability (Vandevijvere et al, 2009). Three interviews over three separate days (two week days and one weekend day) was considered sufficient to account for any intra-individual variability (Lee and Nieman 2007).

3.6.3 URINE SAMPLE COLLECTION AND ANALYSIS

The bespoke home urine collection-kit had been previously validated by researchers at IBERS, AU (Lloyd et al., In Press). A Pro-curo database enabled use of sample code stickers and recorded the location of each sample throughout the study duration for traceability purposes. Consumables were delivered to LJMU and the sampling kits were prepared on site by the principal investigator and encompassed 6 test tubes, 3 transfer straws, blue Safebox, beaker, 6 absorbent pockets, sealable bag, 3 food records, 2 consent forms and blue pre-paid envelope (refer to later Fig. 6.3 in Chapter 6). In accordance to each 24 h MPR, participants collected a morning void urine sample in the sterile beaker and used a transfer straw to decant 4ml of urine into two vacutainers. Participants stored samples in the fridge until returning all samples in a royal mail approved Safebox, along with a completed 3-day food record, 3 used transfer straws and written informed consent form.

3.6.4 ACCEPTABILITY QUESTIONNAIRE

On completion of the 3 x 24 h MPR and urine sample collection, each participant received an invitation via text to participate in an online questionnaire regarding the acceptability of the bespoke urine sampling kit. The questionnaire, uploaded on Online surveys (Jisc, UK), provided participant information with implied consent. The survey asked participants to rate their opinion towards a statement concerning the method on a scale from strong agreement to strong disagreement. Responses were correlated with demographic data from Study 2.

3.7 STUDY DATA ANALYSIS

Data was analysed using IBM SPSS® 24.0 (SPSS Inc. Chicago, USA) and Microsoft Excel 2013. Data screening and cleaning for outliers and errors on the categorical and continuous variables was completed and normality testing using inspection of graphs and Shapiro-Wilk was used prior to any other statistical tests; if the assumption of the normality had not been violated the Sig. was p = < 0.05. Descriptive statistics were computed for socio-demographic, lifestyle, and frequency of takeaway food intake variables for all three studies. PCA, Pearson's, Spearman's, Man Whitney U, Kruskal Wallis H and multivariate analysis of variance (MANOVA) were the statistical tests used to investigate dietary patterns and associations between other variables (takeaway food consumption, socio-demographic and lifestyle factors) in this project. Further detail is provided within the relevant chapters.

Chapter 4
Study 1

4 Chapter 4: Takeaway food consumption, wider dietary patterns and associated determinants: A pilot study

Abstract

Background: Takeaway foods have become increasingly popular and are thought to be associated with a number of non-communicable diseases owing to their high fat, salt and sugar content. Dietary patterns may be more predictive of disease risk than individual foods or nutrients due to the synergistic effects of food combinations. Literature searches have demonstrated that there are no previous studies in the UK that have investigated wider dietary patterns in relation to takeaway foods. ***Aim of Study 1:*** To pilot a dietary survey assessing habitual and takeaway food consumption and define possible dietary patterns of a sample of Liverpool adults in preparation for carrying out a larger scale study on the dietary patterns and socio-demographic and lifestyle factors of takeaway food consumers. ***Method:*** A modified version of the European Prospective Investigation of Cancer (EPIC)-Norfolk food frequency questionnaire (FFQ), to include takeaway food consumption and socio-demographics, was created, tested and optimised in a pilot study (n = 26). Data from the FFQ was converted into portions per week and principal component analysis (PCA) was performed to identify dietary patterns in the study sample. A series of three 24 hour 'Multiple Pass' dietary recalls (3 x 24 h MPR) were carried out and converted into grams per day to validate results from the FFQ. ***Results:*** PCA identified three dietary patterns (characterised as 'Traditional, 'Cosmopolitan' and ''Convenience') which explained 42.9% of the variance in dietary intake. Interestingly, elements of the diet that tracked together in the 'Convenience' pattern included unhealthy

habitual foods (refined grains, cakes, pastries etc.) and takeaway food (Chinese, Indian, kebabs etc.). ***Conclusion:*** Pilot testing showed that PCA is an appropriate technique for dietary pattern analysis, generating three distinct dietary patterns. These findings supported the opening of the main study to use the same techniques on a sample of Merseyside takeaway food consumers.

4.1 CHAPTER OUTLINE

This was a pilot cross-sectional (observational) study to test methods and receive participant feedback on study design. Dietary intake was analysed using two forms of dietary assessment (food frequency questionnaire (FFQ) and three 24 hour 'Multiple Pass' dietary recall (3 x 24 h MPR)). A vital role of the pilot stage was the development of a suitable dietary survey to assess takeaway and habitual food consumption and to compare collection methods. The combined use of two dietary assessment tools was to create several layers of analysis for validation purposes and to collect complementary data. The FFQ focussed on frequency of consumption across a 1-year period and the 3 x 24 h MPR provided detailed information regarding food and drink intake for 3 days in a 1-week period. Takeaway food included those purchased from small, independent outlets (Jaworowska et al., 2014) as well as national/multinational fast-food chains (such as McDonald's, KFC, Domino's Pizza and Subway). Dependant variables, self-reported demographics such as age, gender, ethnicity and lifestyle factors such as physical activity and smoking status, determined associations with dietary patterns produced from principal component analysis (PCA).

4.2 Aims and objectives

Aim: To trial and pilot a two stage dietary survey on a sample of the Liverpool population in preparation for carrying out a larger scale takeaway food study.

Objectives:

1. Test the acceptability of a modified version of the European Prospective Investigation of Cancer (EPIC)-Norfolk FFQ including a detailed section on takeaway foods and background information (including socio-demographics, anthropometrics and lifestyle factors) in preparation of a larger study (Study 2 and 3).
2. Gather preliminary data (habitual diet and takeaway food intake) to explain takeaway food habits and to inform Study 2.
3. Identify dietary patterns of the pilot cohort, as well as explore associations with socio-demographic and lifestyle factors.
4. Evaluate differences between face-to-face and telephone 24 h MPR and validate dietary intake from the FFQ.

4.3 Methods

4.3.1 Study population

Twenty-six (17 female and 9 male) university staff and students aged 18–64 years were recruited for the study at LJMU between January and April 2016. As this was a pilot, no formal power calculation was required. Students and staff at LJMU were invited verbally or received an invitation via email to participate

(Appendix 9.11). Participants received an informed consent form (Appendix 9.13) and participant information sheet (Appendix 9.12) in hard copy and/or email which explained the study. Participants had one week to decide if they would like to take part. Inclusion criteria encouraged the age range of 18–64 years (regarded as the working age for adults). Participants were also required to reside in Liverpool due to convenience sampling and to continue on from previous takeaway food research in Merseyside (Jaworowska et al., 2014). In this pilot study, there was no material or monetary reward offered; however, on completion participants were debriefed which included a general idea of how their role had contributed to the field of nutrition research.

4.3.2 Development of a food frequency questionnaire

The 'Eating Habits Survey' (EHS) was created in accordance to the previously validated EPIC-Norfolk semi-quantitative FFQ (Bingham et al., 1997; University of Cambridge, 2014) (Appendix 9.14) and included a modified extensive list of habitual foods from the original FFQ and new sections regarding takeaway foods and background information. The food items/groups and frequency of consumption ranged from 1 (never or less than once per month) to 9 (6 or more times a day). Whilst maintaining the same food grouping and frequency format, the section regarding various types of takeaway foods was designed. The takeaway foods selected for the questionnaire were sourced from menus of the most commonly consumed types of takeaway food in the UK; Italian/Pizza, Chinese, Indian, kebabs, fried chicken, English/fish & chips and American/fast food (Just Eat, 2017). Takeaway foods included those from small independent outlets (Indian, Chinese, Fish and Chip shops etc.) and fast food franchises (McDonald's, Burger King, Domino's Pizza etc.). In addition, questions regarding takeaway food behaviour were included such as location of consumption, reasons for consumption, social interaction etc. The final amendment to the FFQ was the addition of background information including self-reported socio-demographics (such as marital status and occupation), anthropometric measurements (such as height and weight) and lifestyle factors (such as smoking status and exercise level) (Appendix 9.14).

For background information, demographics such as age, gender, education, ethnic background, adults and children in household etc. were included. Lifestyle factors such as physical activity, supplement use, cigarette smoking and alcohol consumption were also included to investigate associations among these variables and takeaway food consumption. Habitual physical activity was assessed using questions from a Physical Activity Questionnaire (PAQ) adapted from that of Fave et al. (2011) and originally Wareham et al. (2003) as part of the EPIC study. The questions included detailed self-reports of time and intensity levels of occupational and domestic activities. For daily occupational activity, participants selected one of four categories, as in Table 4.1, and for weekly leisure activities (mildly energetic, moderately energetic and vigorous) (Table 4.1) participants recorded time spent (hours per week) on each activity. For leisure activities, total hours for each activity were combined into two variables; low physical activity (LPA) and moderate and vigorous physical activity (MVPA). The duration of leisure activities were truncated; 16 hours for low physical activity, 10 hours for moderate physical activity, and 4 hours for vigorous physical activity. This was to control for any over-estimation of physical activity which may cause misclassification, in accordance to methods used in the NDNS (Mindell, 2014; Bates et al., 2014).

Table 4-1 Occupational daily activity and weekly domestic activity (Wareham et al., 2003)

Domain	Activity	Description in questionnaire
Occupational	**Sedentary**	I am usually sitting and do not walk about much (e.g. in an office)
	Standing	I stand or walk about quite a lot, but do not have to carry or lift things very often (e.g. shop assistant, hairdresser)
	Manual	I usually lift or carry light loads or have to climb stairs or hills often (e.g. plumber, electrician, carpenter)
	Heavy manual	I do heavy work or carry heavy loads often (e.g. dock worker, miner, bricklayer, construction worker)
Leisure/Domestic	**Mildly energetic**	(e.g. walking, gardening, playing darts, general housework)
	Moderately energetic	(e.g. heavy housework or gardening, dancing, golf, cycling, leisurely swimming)
	Vigorous	(e.g. running, competitive swimming or cycling, tennis, football, squash, aerobics)

Self-reported anthropometrics including height, weight and waist circumference were included to determine BMI and Waist-to-height ratio (WHtR) of participants. BMI was calculated using self-reported height and body weight measurements and the equation: Weight (kg)/Height (m^2) = BMI. The BMI classification system determines if individuals are underweight, normal/healthy weight, overweight/pre-obesity, obesity 1, obesity 2, obesity 3 (Table 4.2) according the World Health Organization and NICE guidelines (National Institute of Health and Care Excellence, 2014; World Health Organization, 2016). These cut-offs are designed to correlate with risk of health problems within a population.

Table 4-2 BMI classification

BMI (kg/m^{2})	Category
< 18.5	Underweight
18.5–24.9	Healthy/Normal weight
25.0–29.9	Overweight/Pre-obese
30.0–34.9	Obesity class 1
35.0–39.9	Obesity class 2
≥ 40.0	Obesity class 3

Waist-to-height ratio (WHtR) calculated abdominal obesity using the equation; waist divided by height in meters. The cut off for predicting whole body obesity was 0.53 in men and 0.54 in women using the WHtR ratio; while a ratio of 0.59 and above indicated abdominal obesity in both genders (Browning et al., 2010). However, the risk of developing diabetes and CVD begins to rise with a WHtR above 0.50.

The advantages of using an FFQ as the primary dietary assessment technique included low respondent burden (10–20 minutes to complete), ease of administration (online), low cost and relatively low researcher burden; all of which were important aspects when targeting a large sample (n = 2000) needed for the main study. The reproducibility and validity of the EPIC-Norfolk FFQ has been reported previously (Kroke et al., 1999; Brunner et al., 2001) in addition to dietary patterns defined by factor analysis with data from an FFQ (Hu et al., 1999; Khani et al., 2004; Nanri et al., 2012). The EPIC-Norfolk FFQ had been designed for use on a UK population and therefore suitable for use on a sample of the Liverpool population. However, the original FFQ did not investigate takeaway food consumption and due to this modification, validity and reproducibility may have been affected. Therefore, this study assessed content validity of the FFQ during the development stage by the involvement of other nutrition and health professionals and included informal feedback on the suitability of the questionnaire. Convergent validity was also assessed by using two retrospective methods of dietary assessment (FFQ and 3 x 24 h MPR) that were theoretically similar to one another; 24 h MPR data was collected to validate the FFQ results.

The first stage of the pilot study involved the development of the FFQ based on participant feedback. Whilst completing the FFQ, participants provided comments on difficult questions or recommendations for improvement to the questionnaire design; the reviewed suggestions helped tailor the questionnaire. The takeaway foods section was trialled in three formats (full list, food groups and short list) and the food groups were selected for use as they corresponded well with the style of the EPIC habitual diet section. The addition of images and editing of colour and font size improved the appearance of the questionnaire. Physical activity was also broken down into occupational and leisure activities adapted from the EPIC study (Fave et al., 2011). Finally, the FFQ was renamed the 'Eating Habits Survey' to be understood by a lay audience and due to the inclusion of questions relating to dietary behaviours and background information.

4.3.3 24 HOUR 'MULTIPLE PASS' DIETARY RECALLS (24 H MPR)

Three 24 h MPR (Appendix 9.15) were performed for each participant and involved using a hierarchically structured approach to progressively ask more specific questions during each 'pass'. This was similar to the stages followed in the UK Low Income and Diet Nutrition Survey study (Nelson et al, 2007) and US

Department of Agriculture (USDA) "5-Step Multiple-Pass Approach" (Raper et al., 2004). The initial step recorded date, day of the week and a quick list of all food and drinks consumed in chronological order without interruption. The second step involved a review of all food and drinks recorded (in chronological order), allowing for the addition of forgotten foods and clarification of any ambiguities regarding the type of food or drink consumed. The third step asked questions on meal occasion/time, location, alcohol consumption, supplement/medication use and a checklist of commonly consumed foods and takeaway foods. The fourth step asked detailed food questions including portion size, type, brand, cooking method, foods in combination (e.g. tea with sugar and milk) and any leftovers or second helpings. Finally, step five involved a final probe for any missing information, a question on takeaway food consumption for the previous 7 days and if diet had changed since completing the FFQ. Previous research validating 24 h dietary recalls by direct observation found that it accurately assessed intakes of energy, protein, carbohydrate, and fat in a population of men regardless of their BMI (Conway et al., 2004). Thus, this technique was included as a more accurate measure of dietary intake than the FFQ.

Face-to-face and telephone interview methods of the 24 h MPR were tested against one another for reliability and to determine if one method outperformed another. Once data was collected the 3 x 24 h MPR was analysed; food portions were converted from household measures/approximate servings into grams using a food atlas (Ministry of Agriculture Fisheries and Food, 1993; Nelson et al., 1997). Food items and grams were then added to a data matrix containing the same list of food groups used for the FFQ analysis (n = 18). A sum for each food group was generated and divided by 3 (for 3 x 24 h MPR) to produce an average consumption of each food group in grams/day. These figures validated findings from the FFQ by grouping participants based on their dietary pattern and comparing overall grams/day of specific food groups. This type of validation is convergent validity; the 3 x 24 h MPR was the reference method for validity analysis, similar to previous research that collected data from twelve 24 h dietary recalls to validate a FFQ (Funtikova et al., 2015).

4.3.3.1 Statistical analysis

Statistical analyses were performed using IBM SPSS® 24.0 (SPSS Inc., Chicago, Illinois, USA) and Microsoft Excel 2013. Data screening and cleaning was conducted to check for any outliers and errors using boxplots for categorical (age, gender, education etc.) and continuous (BMI, WHtR, LPA etc.) variables. Normality was assessed using inspection of graphs and statistical test (Shapiro-Wilk) prior to any other tests; if the assumption of the normality had not been violated the significance was $p > 0.05$. Descriptive statistics explained socio-demographic, lifestyle, frequency of takeaway food intake and takeaway food behaviour.

4.3.3.2 Dietary pattern Analysis by Principal Component Analysis

Dietary pattern analysis, as a more holistic approach to dietary assessment, has become popular over the past two decades. There are a number of ways to conduct dietary pattern analysis (Appendix 9.4), however, there are two general overarching approaches; *a priori* which is a hypothesis-driven approach, and *a posteriori* which is purely data-driven (Granic et al., 2016). Each method is built on evidence and includes some degree of subjectivity (Moeller et al., 2007). Factor analysis encompasses a complex family of data-driven statistical techniques (Pallant, 2013) such as principal component analysis (PCA) (Smith et al., 2013). PCA reduces the original large set of correlated dietary variables into a new, smaller set of uncorrelated variables, which captures the major dietary traits in a population (Bamia et al., 2005; Reedy et al., 2010). PCA simplifies data interpretation by emphasizing variation to draw out strong patterns. Several studies have used PCA to identify the dietary patterns of populations in relation to health risk, socio-demographic correlates and other variables such as cognitive status and lifestyle (Appendix 9.4). However, no previous studies have explored takeaway food consumption in relation to wider dietary patterns using PCA. This technique may be able to identify correlations between foods and food groups from a myriad of nutritional data (Mullie et al., 2010). PCA selected for this project was a more sophisticated technique than the methods used in previous studies on takeaway food that generally assess frequency of consumption. This technique aimed to deliver a deeper understanding of habitual dietary patterns and consumption of takeaway foods in combination with other foods in the diet.

To perform PCA, a number of decisions occur during the analytical process, including how to quantify the food groups (input variables) (Smith et al., 2013). Thus, the input variables used in PCA vary across studies (Newby and Tucker, 2004) and include frequency of consumption, gram weights, energy adjusted weight, daily percentage energy contribution and binary variables (Smith et al., 2013). Selected frequency of consumption for each food in the current FFQ was converted to portions per week for dietary pattern analysis as follows: NEVER or less than once/month = 0, 1–3 per month = 0.5, once a week = 1, 2–4 per week = 3, 5–6 per week = 5.5, once a day = 7, 2–3 per day = 17.5, 4–5 per day = 31.5, 6+ per day = 56 (calculation based on 8 servings per day); similar methods have been reported previously (Smith et al., 2011). The number of input variables defined are study specific and can vary according to sample size and research question (Appendix 9.4). Two methods classified the 212 individual food items before applying PCA. In the first instance, the food and drink items were combined and collapsed into 28 food groups (first column in Table 4.3) and into 18 food groups (second column in Table 4.3), respectively, with similar nutrient profiles. This process was carried out in line with commonly consumed foods and for foods of key interest such takeaway food groups. Previous studies have aggregated foods into groups based on their nutrient profiles (Pryer et al., 2001; Haveman-Nies et al., 2001; Hearty and Gibney, 2009; Ashby-Mitchell et al., 2015).

PCA performed in SPSS, with the procedure ‘dimension reduction’ and ‘FACTOR’ on both sets of food groups identified the primary components, which accounted for variation in dietary intake. In the current study, the smaller set of food groups (n = 18) was deemed preferable due to the small sample size (n = 26) (Pallant, 2013). Eight principal components had eigenvalues > 1 (classifying them as the initial major components according to Kaiser’s criterion); however, a graphical evaluation of the scree plot showed a clear break after the third component (Appendix 9.18) (Pallant, 2013). Therefore, it was decided to retain three components. A high positive factor loading for a given food item/group indicated high intake within that food pattern, and a high negative factor loading indicated low intake of those food items/groups. Interpretation of the data and earlier literature helped to label the derived components (dietary patterns) (Barker et al., 1990; Pryer et al., 2001; McKeever et al., 2010).

Table 4-3 Food grouping used in the dietary pattern analysis of the pilot cohort

Food Group 1	Food Group 2	Definition and content
1. Red meat **2. Poultry** **3. Processed meats** **4. Offal**	**1. Meat**	Beef, beef burgers (not takeaway), pork, lamb, liver, bacon, ham, processed meats, sausages, pies, chicken, meat soup
5. Fish and seafood	**2. Fish and seafood**	Fried fish, breaded fish, white fish, oily fish, shellfish
6. Refined grains	**3. Refined grains**	White bread, scones, crackers, pitta, garlic bread, sugary cereal, plain cereal, white rice, pasta, tinned pasta, noodles, Yorkshire puddings, lasagne, pizza (not takeaway)
7. Wholegrains/ meal	**4. Wholegrains/ meal**	Brown bread, wholemeal bread, porridge, all bran, wholegrain cereal, brown rice, wholemeal pasta
8. Potatoes	**5. Potatoes**	Boiled potatoes, potato salad, chips, roast potatoes
9. Full fat dairy **10. Low fat dairy**	**6. Dairy**	Single cream, double cream, full fat yoghurt, dairy dessert, coffee whitener, cheese, low fat yoghurt, cottage cheese, dairy milk
11. Eggs	**7. Eggs**	Boiled, scrambled, fried, poached, quiche
12. Fats and oils	**8. Fats and oils**	Butter, margarine, polyunsaturated margarine, other margarine, low fat spread, mayonnaise, low calorie mayonnaise, French dressing, salad dressing
13. Cakes/pastry /crisps	**9. Cakes/pastry /crisps**	Chocolate biscuits, plain biscuits, cakes, buns, sweet pies, crisps
14. Sweets and desserts	**10. Sweets and desserts**	Milk puddings, ice cream, chocolate, sweets, sugar
15. Sauces and condiments	**11. Sauces and condiments**	Tomato sauces, ketchup, other sauces, relishes, marmite, dips, jam, peanut butter, chocolate spread
16. Tea and coffee	**12. Tea and coffee**	Tea and Coffee
17. Alcohol	**13. Alcohol**	Wine, beer, liquors, spirits
18. SSB	**14. SSB**	Low calorie SSB, cocoa, Horlicks, SSB, pure fruit juice, cordial/squash

19. Fruit	**15. Fruit**	Apples, pears, oranges, grapefruit, bananas, grapes, melon, peaches, strawberries, avocado, tinned fruit, dried fruit
20. Vegetables **21. Soup**	**16. Vegetables**	Carrots, spinach, broccoli, brussel sprouts, cabbage, peas, green beans, courgettes, cauliflower, parsnips, leeks, onions, garlic, mushrooms, sweet peppers, beansprouts, green salad, mixed vegetables, watercress, tomatoes, sweetcorn, beetroot, coleslaw, vegetable soup
22. Legumes and nuts	**17. Legumes and nuts**	Bakes beans, pulses, tofu, nuts, milk from plant source (Soya, Almond)
23. Fast Food	**18. Takeaway Food**	Beef burger, chicken burger, fried chicken, fish burger, veggie burger, mozzarella sticks, fries, McDonald's breakfast, Subway, sauces and dips
24. Chinese		Chicken wings, spare ribs, crispy duck, spring rolls, prawn crackers, siu mai, skewered chicken, soup, salt and pepper chicken, peking dish, lemon/orange/plum sauce dish, satay dish, sweet and sour, curry, chop suey, kung po, foo yung, chow mein, beef dish, chicken or Duck Veg, king prawn, fried rice, salt and pepper chips, noodles, boiled rice, chinese sauces
25. Indian		Poppadums, samosa, tikka starter, tandoori, korma, tikka masala, rogan josh, bhuna, biryani, balti, sagwala, tikka jalfraizi, dupiaza, medium curry, achari, vindaloo, pathia, pasanda, special, butter chicken, saag, dal, naan, paratha, pilau, fried rice, boiled rice
26. English		Fish, scampi, sausage, omelette, roast chicken, chips, sauces
27. Pizza		Cheese, meat, seafood, vegetarian, garlic bread
28. Kebab		Donner, chicken, sheesh

PCA calculates unique component scores for each participant by determining the regression weights, multiplying each participant response by the respective weights and then summing the products. The resulting sum (component score) for each participant indicates the extent to which their diet conformed to one of the retained components (dietary patterns). Each score had a mean of 0, this ‘centring’ of the data is necessary when performing PCA to standardise the data. The further away the score is from the mean, the more or less likely a participant adhered to a dietary pattern, with a higher score indicating closer adherence to that dietary pattern. These values were used as dependent variables for group comparisons.

4.3.4 ASSOCIATIONS WITH DEMOGRAPHIC AND LIFESTYLE FACTORS

Component scores for each dietary pattern were produced via PCA and were used to explore associations with other lifestyle variables or differences between socio-demographic groups. The demographic and lifestyle covariates included in the analysis were similar to previous research (Batis et al., 2016). With no violations to normality; Pearson’s correlation (Table 4.4) was used to determine if the dietary patterns were associated with any of the continuous variables (BMI, WHtR, physical activity). Multivariate analysis of variance (MANOVA) investigated differences between two or more groups of categorical data (age group, gender, education level, smoking status, supplement use and occupational physical activity) with continuous data (dietary patterns).

Table 4-4 Pearson’s correlation guidelines (Cohen, 1988)

Coefficient Value	Strength of Association
0.1 < | *r* | < 0.3	small correlation
0.3 < | *r* | < 0.5	medium/moderate correlation
| *r* | > 0.5	large/strong correlation

4.4 RESULTS

4.4.1 PARTICIPANT CHARACTERISTICS

The total sample was 26 (17 female, 9 males) participants from LJMU (Table 4.5) with a mean BMI of 23.64 Kg/m² (Table 4.6). No exclusions were made as all participants (n = 26) had provided consent and completed the FFQ. The initial ten participants also provided feedback on the FFQ design. Nineteen participants, from the FFQ sample, completed 3 x 24 h MPR.

Table 4-5 Study population socio-demographic characteristics 2016 (n = 26)

Variable	**Level**	***n***	**%**
Age group (years)	18 – 25	5	19.2
	26 – 35	9	34.6
	36 – 45	4	15.4
	46 – 55	4	15.4
	56 – 64	4	15.4
Gender	Female	17	34.6
	Male	9	65.4
Ethnicity	White British	24	92.3
	Other	2	7.7
Marital status	Single	13	50.0
	Married (including civil partnership)	11	42.3
	Divorced/Separated/Widowed	2	7.7
Living arrangements	On own	3	11.5
	Wife/Husband/Partner	15	57.7
	Parents	4	15.4
	Friends	3	11.5
	Children	1	3.8
Adults in household (18+)	1	4	15.4
	2	14	53.8
	3	3	11.5
	4 or more	5	19.2

Children in household (U18)	0	14	53.8
	1	5	19.2
	2	3	11.5
	3	1	3.8
	4 or more	1	3.8
Educational achievement	No formal qualifications	0	0
	GCSE/O-Level	0	0
	A-Level or Equivalent	4	15.4
	Degree Level	9	34.6
	Postgraduate Qualification	13	50.0
NS-SEC	Managerial/Professional	8	30.8
	Intermediate	5	19.2
	Routine/Manual	3	11.5
	Student/Unemployed/Retired	10	38.5

NB: n = number of participants. NS-SEC = National Statistics Socio-economic classification. U18 = Under 18 years of age.

Table 4-6 Study population lifestyle and self-reported anthropometric characteristics 2016 (n = 26)

Variable	Level	*n*	%	Mean (SD)
BMI Kg/m²		26		23.64 (3.38)
WHtR		26		0.47 (0.06)
Alcohol drinks/week		26		4.31 (5.38)
Leisure activity	LPA	26		9.28 (6.44)
	MVPA	26		4.62 (3.75)
Currently smoking	Yes	4	15.4	
	No	22	84.6	
Occupational activity	Sedentary	11	42.3	
	Standing	11	42.3	
	Manual	4	15.4	
	Heavy manual	0		
Supplements	Yes	12	46.2	
	No	14	53.8	

NB: n = number of participants. SD = Standard deviation. PA = Physical activity. WHtR = Waist-to-height ratio. LPA = Low physical activity. MVPA = Moderate and vigorous physical activity.

4.4.2 TAKEAWAY FOOD CONSUMPTION

Results from the FFQ showed that the majority (42%) of the pilot sample consumed takeaway food less than once a month, 35% 1–3 times per month, 23% once a week or more (Fig. 4.1).

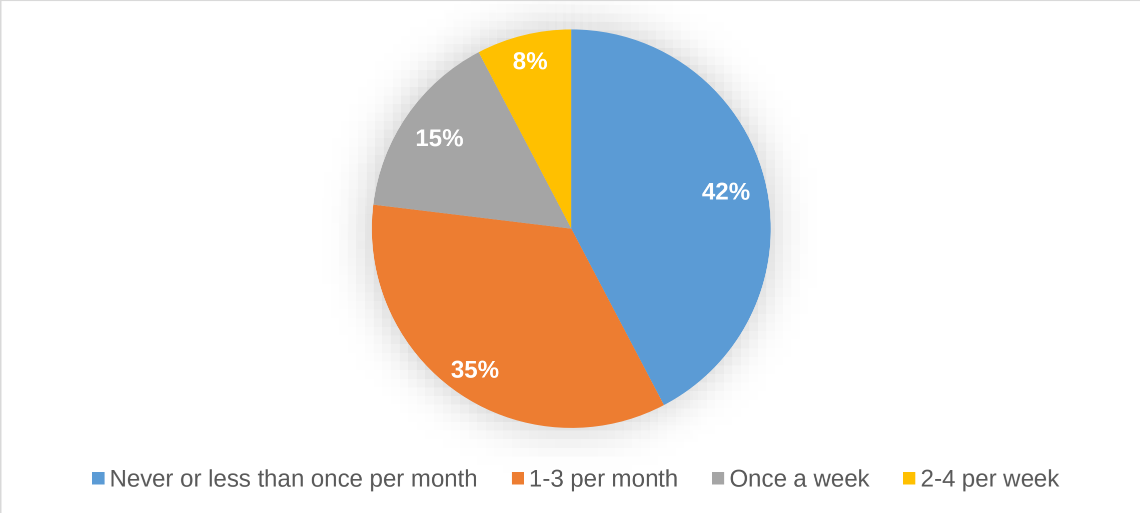


Figure 4-1 How often do you usually eat takeaway/fast food? (n = 26)

There were some clear differences in the types of takeaway food consumed, with Chinese being most popular (n = 12), followed by pizza (n = 10), fish and chips (n = 9) and Indian (n = 8) respectively (Fig. 4.2). Other was 'chicken burger'.

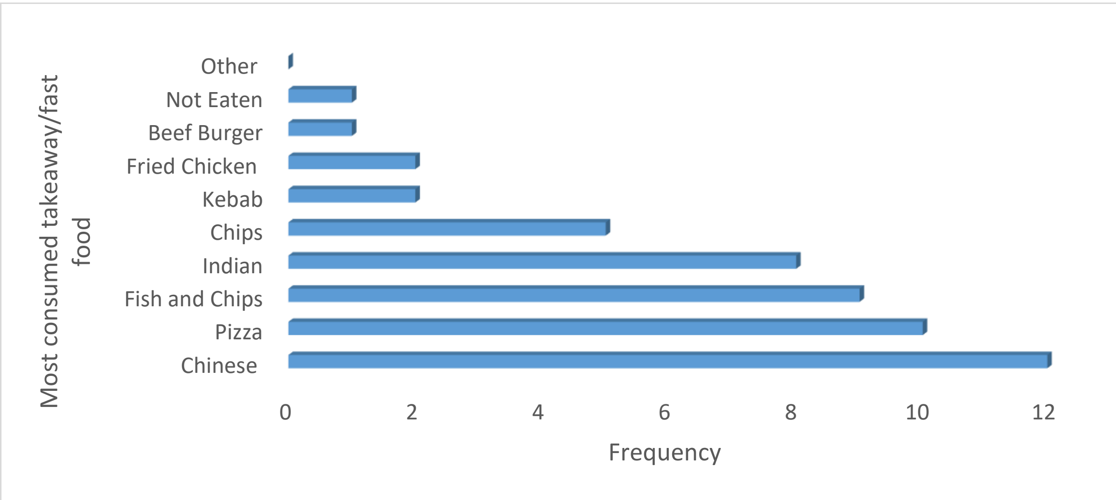


Figure 4-2 What type of takeaway/fast food do you eat most often? (Please tick all that apply) (n = 26)

There were a range of motives expressed by participants for takeaway food consumption, with ‘taste’ (n = 13) and ‘social occasions’ (n = 13) leading the results (Fig. 4.3). Other prominent reasons for consumption were that takeaways ‘are easily available’ (n = 12), ‘my friends/family like it’ (n= 11), ‘available close to the home’ (n = 8) and ‘too busy to cook’ (n = 8). Some other reasons described by participants included ‘for convenience’ and as a ‘treat at the weekend’.

Figure 4-3 Why do you eat takeaway/fast food? (Please tick all that apply) (n = 26)

Participants mostly consumed takeaway food with family (n = 19) and friends (n = 15) (Fig. 4.4). Other was partner.

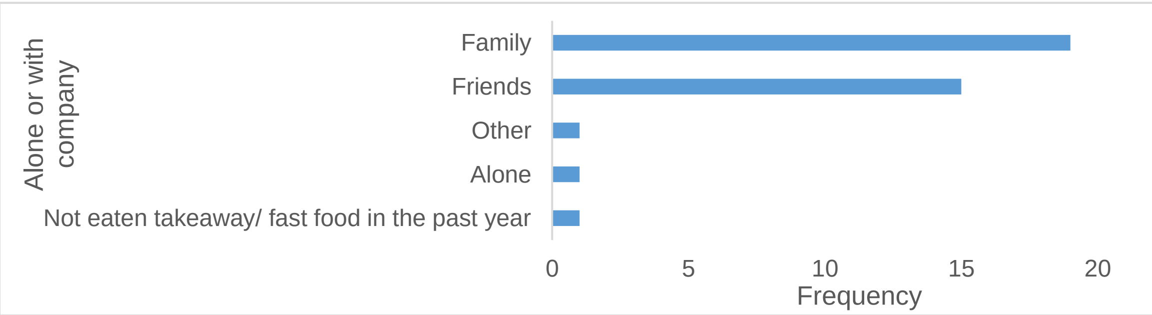


Figure 4-4 Who do you eat takeaway/fast food with? (Please tick all that apply) (n = 26)

The majority of participants consumed takeaway food at home (n = 24) (Fig. 4.5). Some indicated consumption at friends or with families (n = 9) and very few at the takeaway food outlet (n = 1).

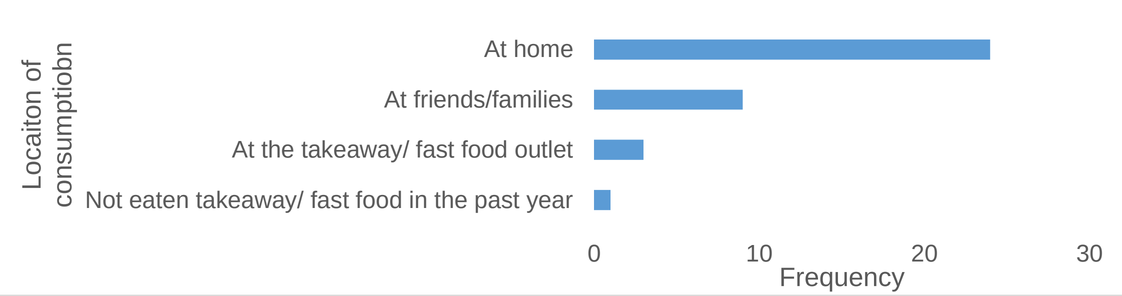


Figure 4-5 Where do you eat takeaway/fast food? (Please tick all that apply) (n = 26)

4.4.3 Dietary patterns from FFQ data

The 18 food groups from the FFQ dietary assessment were subjected to PCA. Prior to performing PCA, the suitability of data factor analysis was assessed. Inspection of the correlation matrix (Appendix 9.17) revealed the presence of many coefficients of 0.3 and above. The Kaiser-Meyer-Olkin value was 0.329, lower than the recommended value of > 0.5 (Kaiser, 1974) however, this could be explained by the small sample size. The Barlett's Test of Sphericity (Bartlett, 1954) reached statistical significance, supporting the factorability of the correlation matrix.

PCA revealed the presence of eight components with eigenvalues exceeding 1, explaining 17.0%, 13.9%, 12.0%, 11.9%, 8.6%, 7.4%, 6.4% and 5.9% of the variance respectively. However, inspection of the scree plot (Appendix 9.18) revealed a clear break after the third component. Therefore, three components were retained as instructed by Pallant (2013) and these factors represented a cumulative percentage of 42.9% of the inter-individual variability.

An oblimin rotation aided the interpretation of these three components. The rotated solution revealed the presence of a number of high positive and high negative factor loadings (Table 4.7).

The first component in the pattern coefficients, described as a ‘Traditional’ dietary pattern, had high positive loadings obtained for potatoes (0.536), alcohol (0.520), meat (0.490), and sweets and desserts (0.360). Negative loadings were obtained for vegetables (−0.715), eggs (−0.567), legumes and nuts (−0.492), fruit (−0.458), sauces and condiments (−0.425), fish and seafood (−0.372) and takeaway food (−0.370) (Table 4.7).

For the second component, it was evident that the high positive loadings included tea and coffee (0.626), meat (0.501), fish and seafood (0.358), and eggs (0.305) and high negative loadings for SSB (−0.784), cakes, pastry and crisps (−0.618), legumes and nuts (−0.409), and wholegrains/meal (−0.398) (Table 4.7). Thus, this pattern was described as ‘Cosmopolitan’.

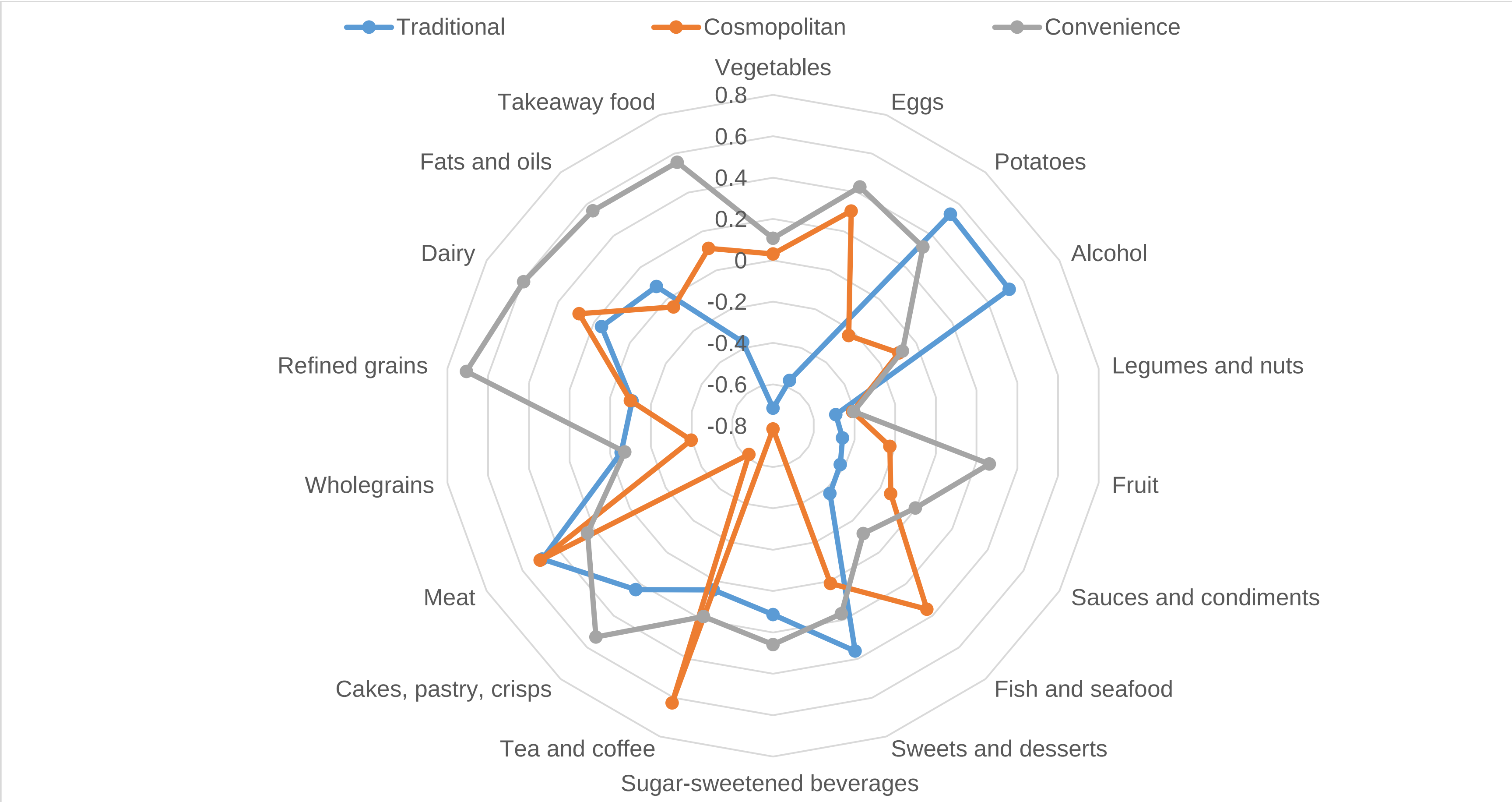
The third component was characterised by high positive loadings for ‘convenience’ food including refined grains (0.707), dairy (0.593), fats and oils (0.557), takeaway food (0.556), cakes, pastry and crisps (0.533), eggs (0.428), and potatoes (0.329) (Table 4.7) and high negative loadings for legumes and nuts (−0.404). This pattern was labelled ‘Convenience’.

A visual representation of the three dietary patterns shows the variance between the diets of the study sample (Fig. 4.6).

Table 4-7 Rotated component loadings derived from PCA with Oblimin rotation for dietary variables that constitute the three main dietary patterns identified

Food Group	Principal Component (Dietary Pattern)			Communalities
	C 1	C 2	C 3	
Vegetables	−0.715	0.031	0.106	0.510
Eggs	−0.567	0.305	0.428	0.568
Potatoes	0.536	−0.231	0.329	0.478
Alcohol	0.520	−0.097	−0.077	0.280
Legumes and nuts	−0.492	−0.409	−0.404	0.622
Fruit	−0.458	−0.226	0.263	0.299
Sauces and condiments	−0.425	−0.142	−0.004	0.200
Fish and seafood	−0.372	0.358	−0.120	0.289
Sweets and desserts	0.360	0.012	0.168	0.169
SSB	0.113	−0.784	0.258	0.681
Tea and coffee	0.045	0.626	0.182	0.439
Cakes, pastry, crisps	0.234	−0.618	0.533	0.715
Meat	0.490	0.501	0.237	0.576
Wholegrains/meal	−0.054	−0.398	−0.072	0.170
Refined grains	−0.107	−0.099	0.707	0.500
Dairy	0.158	0.283	0.593	0.490
Fats and oils	0.078	−0.050	0.557	0.325
Takeaway food	−0.370	0.113	0.556	0.427

NB: Major loadings for each item are in red (factor loadings > 0.3 or < −0.3 are considered significant). Extraction method: Principal component analysis. Rotation method: Oblimin with Kaiser Normalisation. Communalities represent the correlations among the factors.



NB: Blue line = the 'Traditional' dietary pattern (DP1); orange line = the 'Cosmopolitan' pattern (DP2); grey line = the 'Convenience' dietary pattern (DP3).

Figure 4-6 Radar graph of dietary patterns and factor loadings derived through PCA on 18 FFQ food groups (n = 26)

4.4.4 DEMOGRAPHICS IN RELATION TO DIETARY PATTERNS

A Pearson's product-moment correlation assessed the relationship between the Traditional dietary pattern (DP1), Cosmopolitan dietary pattern (DP2), Convenience dietary pattern (DP3), BMI, WHtR, low physical activity (LPA), and moderate and vigorous physical activity (MVPA). Shapiro-Wilk's test assessed the normal distribution of all variables ($p > 0.05$). There was a moderate positive correlation between Cosmopolitan DP2 and low physical activity ($p < 0.05$). Moderate and vigorous physical activity was negatively correlated with WHtR ($p < 0.05$). There was a strong positive correlation between BMI and WHtR ($p < 0.005$). All other correlations were non-significant (Table 4.8).

Table 4-8 Pearson's correlations on FFQ dietary patterns

	BMI	DP1	DP2	DP3	LPA	MVPA
DP1	−0.60					
DP2	0.13	−0.01				
DP3	−0.21	0.09	0.05			
LPA	0.28	−0.08	0.40*	0.11		
MVPA	−0.34	−0.12	0.03	−0.12	−0.18	
WHtR	0.78**	0.00	0.12	0.10	0.22	−0.42*

NB: Significant correlations highlighted in red. **Correlation is significant at the 0.01 level (2-tailed). *Correlation is significant at the 0.05 level (2-tailed).

One-way between-groups (Traditional, Cosmopolitan, and Convenience dietary scores respectively) multivariate analysis of variance investigated gender differences in dietary patterns. Preliminary assumption testing to check for normality, linearity, univariate and multivariate outliers, homogeneity of variancecovariance matrices, and multicollinearity noted no serious violations. There was no significant difference between males and females on the combined dependant variables (Table 4.9). The same methods (one-way analysis of variance (ANOVA)) tested for differences between groups based on education level, smoking status, occupational physical activity and supplements. Results showed no significant difference between any groups (Table 4.9).

Table 4-9 Multivariate analysis of variance

	F	P value	Wilks' Lambda	Partial Eta Squared
Gender	(3, 22) = 0.33	0.805	0.96	0.04
Age group	(3, 19) = 1.38	0.208	0.47	0.22
Education	(3, 21) = 0.75	0.616	0.82	0.10
Smoke	(3,22) = 1.03	0.399	0.88	0.12
Supplements	(3,22) = 2.85	0.061	0.72	0.28
Occupational PA	(3,21) = 0.79	0.590	0.80	0.10

NB: PA = Physical activity. P value = statistical sig. < 0.05.

4.4.5 FFQ Validation from 24 h MPR data

Figure 4.7 shows the average quantity consumed (grams/day) from 3 x 24 h MPR for the 18 food groups identified through the dietary pattern analysis of FFQ data, which has been split into the three dietary patterns; 'Traditional', 'Cosmopolitan', and 'Convenience' to validate the FFQ (Fig. 4.6) (Appendix 9.16). The horizontal axis represents the food item/groups and the vertical axis represents the amount consumed in grams/day. Data from the 3 x 24 h MPR has shown that the Traditional dietary pattern group had higher intakes of meat, potatoes, dairy and cakes, biscuits, crisps, sauces, condiments and fruit than the other dietary patterns. The Traditional dietary pattern was lowest in eggs, vegetables, legumes and nuts, and sweets and desserts.

The participants from the FFQ with a Cosmopolitan dietary pattern consumed the highest amounts of legumes, vegetables, fish, tea and coffee, wholegrains/meal, fats, oils and sweet desserts than the Traditional or Convenience patterns in the 24 h MPR. This group consumed less takeaway food, SSB, alcohol, meat, and sauces and condiments.

Finally, within the Convenience dietary pattern more alcohol, SSB, refined grains, takeaways and eggs were reported than the other patterns, and less wholegrains/meal, potatoes, fruit, fish, and fats and oils.

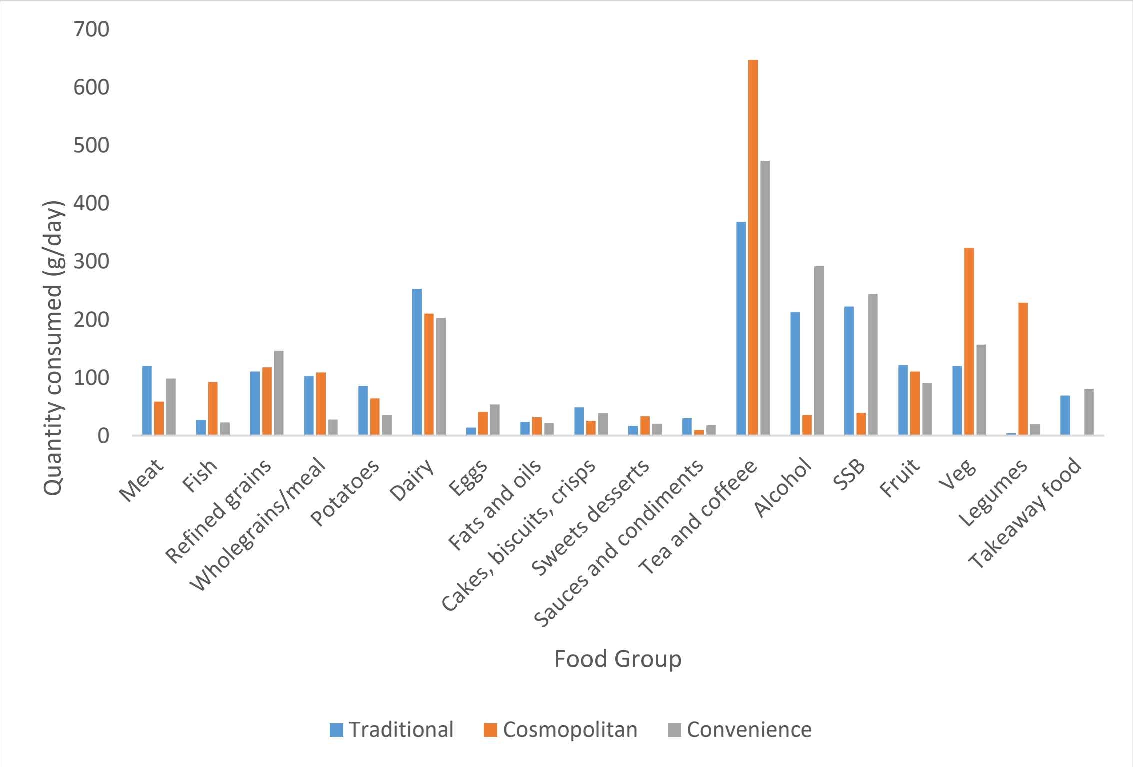


Figure 4-7 Average consumption of 18 food groups in grams/day from 3 x 24 h MPR for three dietary patterns identified using PCA on FFQ data

4.5 Discussion

4.5.1 Takeaway food consumption

The findings from this study showed that 23% of participants consumed takeaway food at least once per week and 35% 1–3 times per month (Fig. 4.1) which was lower than previous findings (Public Health England, 2017b); this may be because of the homogenous sample recruited at LJMU (e.g. educational levels/health literacy might be higher in this group (or less varied) than a more general population) and small sample size. Recent figures have shown that more than one quarter (27.1%) of adults and one fifth of children consume food from out-of-home food outlets at least once a week (Public Health England, 2017a) but the study does not define how many are takeaways. Previous UK data showed that 27% of the population in England consumed foods from takeaway outlets at least once a week (Food Standards Agency, 2014). However, data on consumer purchasing has shown that Liverpool and other areas in the North West have a marked growth in household spending on takeaway food, an increase of 11.8% since 2014 to an average of £30 a month spent by each household on takeaway in 2016 (Just Eat, 2017). Therefore, consumption is likely to have increased during the 4 years since the FSA report. The current study presented some clear differences with the types of takeaway food consumed. Chinese and pizza meals were the most popular (Fig. 4.2), which were associated with habitual convenience foods (Fig. 4.6). In agreement with these results, a national report found that Chinese was reported as the UK's second favourite takeaway food after Italian pizza for 2012, 2014 and 2016 (Just Eat, 2017). Notably, these results may lack academic rigour as a company within the food sector produced them, yet currently there is no academic reference available.

Participant reasons for takeaway food consumption were mainly for ‘taste’ and ‘social occasions’ (Fig. 4.3). Similarly, a study conducted on Australian adults, between the ages of 18 and 45 years, suggested that fast food consumption was influenced by a general demand for meals that were tasty, satisfying and convenient (Dunn et al., 2011). Moreover, Srivastava (2015) stated that some individuals chose fast food restaurants as a way to spend time with friends, family or someone special; this was also apparent in the current study (Fig. 4.4). An additional determinant expressed by the sample in the current study was the increased availability of takeaway food (Fig. 4.4) suggesting strong environmental influences (Townshend and Lake, 2017). Feng et al. (2010), showed that an abundance of unhealthy and energy-dense food in the environment disrupts an individual’s ability to make healthy food choices. Finally, most participants reported takeaway food consumption in the presence of family and friends and at home suggesting a strong social aspect to takeaway food (Fig. 4.4). A recent qualitative study in the North West of England exploring the sociocultural experiences of takeaway food consumers reported similar findings; takeaway food consumption being seen as a social activity in its own right and also being associated with a number of alternative social activities (birthdays, anniversaries, nights out etc.) (Blow et al., 2019). Findings from the current study (Fig. 4.4) and from the literature highlights the significance of socialising over takeaway food and providing the platform to bond with others whilst building relationships. This factor highlights a problem for shifting habits of takeaway consumption due to family/social connections and a feeling of isolation if one individual deviates from the norm, suggesting a potential barrier to change (Roberto et al., 2015). These results also reinforce the concept that obesity is a socially-communicable phenomenon (Allen and Feigl, 2017; Clark, 2017) that requires cultural change to see a dramatic shift in behaviour and health outcomes (Haidar and Cosman, 2011). Therefore, when designing policies and interventions to combat takeaway food consumption and obesity, a family oriented/societal informed approach is necessary.

4.5.2 Dietary patterns of study sample

In a sample of 26 participants, PCA was able to identify three distinct dietary patterns (Fig 4.6), one of which linked to higher takeaway food consumption. The first dietary pattern, accounting for 17.0% of the total variance, was the most identifiable dietary behaviour to emerge from the analysis (Table 4.7). This was characterised as the 'Traditional' dietary pattern in Liverpool reflecting patterns of eating centred on core foods such as potatoes, meat and alcohol (Fig. 4.6). The associations between the traditional diet and drinking habits are worthy of comment. Liverpool had the second highest level of harmful drinking in the North West and the fourth highest in the country between 2014/2015 (Public Health England, 2016). Similar to the findings reported herein, another study identified a drinker/social pattern characterised by the highest factor loadings for white bread, alcohol, fats and meat dishes (McCourt et al., 2014). A study on British adults defined a traditional British diet as one containing white bread/refined cereals, butter and other margarines, tea and sugar/confectionary, moderately high intakes for cakes/pastries, puddings, high fat dairy products and meat ham/bacon, potatoes and vegetables (Pryer et al., 2001). Previous research using PCA found that a dietary pattern high in meat and alcohol and low in micronutrients positively correlated with increasing arterial stiffness (Kesse-Guyot et al., 2010). A more traditional dietary pattern (high intake of meat and potatoes and lower intake of soy and cereal) was associated with adverse effects on lung function and chronic obstructive pulmonary disease in adults from the Netherlands (McKeever et al., 2010). Consumers of the Traditional dietary pattern tended to be smokers however, the data was adjusted for age, sex, age squared, height, age × height, smoking status, pack-years of smoking, BMI, educational level, and location by using linear regression (McKeever et al., 2010).

The second component in the present study, accounting for 13.9% of the total variance, represented high intakes of caffeine from tea and coffee, protein from animal sources such as eggs, meat and fish and low intakes of relatively unhealthy foods such as SSB, cakes, pastry and crisps (Fig. 4.6). Thus, a ‘Cosmopolitan’ dietary pattern described component 2 (Table 4.7). In line with these results, a study on the dietary patterns of British adults described a cluster of females as consuming a “healthier cosmopolitan diet” that encompassed moderate to high intakes of coffee, vegetables, salads, poultry and median intakes for fish (Pryer et al., 2001). McKeever et al. (2010) classified a cosmopolitan dietary patterns as having higher intakes of vegetables, fish, and chicken. A study on the eating patterns of the general Dutch population also labelled a pattern ‘Cosmopolitan’ with greater intakes of fried vegetables, salad, rice, chicken, fish, and wine (van Dam et al., 2003). In the Dutch study, the Cosmopolitan dietary pattern was significantly associated with higher HDL-cholesterol concentrations and lower blood pressure, than the ‘Traditional Dutch’ or ‘Refined-Foods’ patterns (van Dam et al., 2003); reducing the risk of CVD (He et al., 2014; Maranhão and Freitas, 2014). This evidence suggests that the Cosmopolitan dietary pattern is indicative of a healthier pattern and that different dietary patterns may be associated with the risk of non-communicable disease to varying degrees.

The third component was laden with fast and convenience foods high in sugar, fat and salt including refined grains, dairy, fats and oils, takeaway food and cakes, pastry and crisps, accounting for 12.0% of the total variance in the population (Fig. 4.6). Interestingly, the elements of the diet that tracked together in the ‘Convenience’ dietary pattern were unhealthy habitual foods (refined grains, cakes, pastries, crisps etc.) and takeaway food (Table 4.7). These findings support the concept that takeaway food consumption is a marker of an unhealthy diet overall, similar to findings reported by Poti et al. (2014) on US children. The Convenience dietary pattern in this study (Fig. 4.6) was comparable to the ‘Western’ dietary pattern described by Hu et al. (2000) which comprises sweets, desserts, soft drinks, red meat, processed meats and high fat dairy products, with a lower consumption of fish (omega-3 fatty acids), wholegrains, fruit and vegetables. A ‘Western’ dietary pattern was considered to be high in fat and cholesterol, high-protein, high-sugar, and contain excess salt intake (Manzel et al., 2014). PCA derived similar dietary patterns in Iranian adults from FFQ data, including a ‘Traditional’ dietary pattern and ‘Western’ dietary pattern (Hosseinzadeh et al., 2016).

Findings from the current study suggest that takeaway food is merely one component of an overall unhealthy dietary pattern, with habitual dietary patterns laden in energy-dense nutrient-poor foods. Therefore, targeting takeaway food in isolation without addressing other western dietary habits would likely have little impact on health. For example, if takeaway food was no longer available, individuals may replace their calorie intake with other energy-dense nutrient-poor foods including SSBs, sweets, cake, crisps and other processed food.

4.5.3 DIETARY PATTERNS AND ASSOCIATIONS WITH SOCIO-DEMOGRAPHIC AND LIFESTYLE FACTORS

The only significant correlation in the pilot study was that a higher Cosmopolitan dietary pattern score correlated moderately with increased low physical activity (walking, gardening, cleaning etc.) (Table 4.8). Thus, suggesting that a healthier dietary pattern is a marker of other healthier behaviours such as physical activity. Van Dam et al. (2003) carried out a comparable study on the Dutch population and found that higher cosmopolitan-pattern scores were associated with more leisure physical activity, in addition to a higher educational level, light cigarette smoking (< 10 cigarettes/day), and supplement use. Moreover, a recent cross-sectional study on Polish girls (aged 13–21 years) using PCA to produce physical activity and dietary patterns stated that the highest levels of physical activity were associated with higher scores for a fruit and vegetable dietary pattern (Wadolowska et al., 2016).

In the current study, there were no correlations among the Traditional or Convenience dietary patterns with all other socio-demographic or lifestyle variables (Table 4.8; Table 4.9), which may be a result of the small sample size. In contrast, consumption of a Traditional dietary pattern was associated with older age in Dutch adults, and consumption of a Refined-Foods pattern was associated with younger age, but both were associated with lower educational level, cigarette smoking, less physical activity, and BMI (van Dam et al., 2003). Results from studies in the UK have reported that those with a higher socio-economic status and non-manual social classes had healthier dietary patterns (Barker et al., 1990; Pryer et al., 2001). In a study on the very old population, a 'Low Meat' dietary pattern was regarded as favourable as it was associated with better health (least disabled, cognitively impaired, and depressed), increased physical activity and higher socio-economic status than for the other dietary patterns (Granic et al., 2015). Whereas lower educational attainment remained a significant predictor of 'High Red Meat' and 'High Butter' membership compared with 'Low Meat' (Granic et al., 2015). Despite the findings, a host of other variables such as neighbourhood takeaway food exposure, social relations, and cultural norms may influence takeaway/fast food intake (Stok et al., 2014; Burgoine et al., 2016).

4.5.4 FFQ DIETARY PATTERN VALIDATION

Principal component analysis (PCA) could not be performed on the 24 h MPR data due to the smaller sample size (n = 19) therefore, the 18 food groups in the three dietary patterns (Traditional, Cosmopolitan and Convenience) from the FFQ data validated the 18 food groups in 24 h MPR data by calculating grams/day (Fig. 4.7; Appendix 9.16). Findings from the 24 h MPR were concordant with many of the traits defined by the dietary patterns from the FFQ data. Participants scoring highly on the Traditional dietary pattern in the FFQ, characterised by positive factor loadings for meat, potatoes, and alcohol, also consumed the highest amount of meat and potatoes, during the 3 x 24 h MPR (Fig. 4.7).

According to the 24 h MPR data, participants with a Traditional dietary pattern had higher intakes of meat (120 g/day), potatoes (86 g/day), dairy (253 g/day), cakes/biscuits/crisps (49 g/day), sauces/condiments (30 g/day), and fruit (121 g/day) than the other dietary patterns (Appendix 9.16). The FFQ's Traditional dietary pattern presented negative loadings for vegetables, eggs, legumes and nuts, fruit, sauces and condiments, fish and seafood, and takeaway food (Fig. 4.6). The 24 h MPR showed similar results for the Traditional group including the lowest intakes for vegetables (120 g/day), eggs (14 g/day) and legumes and nuts (4 g/day) (Appendix 9.16). However, alcohol consumption was slightly lower in the Traditional dietary pattern group (213 g/day) compared to the Convenience dietary pattern group (292 g/day) (Appendix 9.16). Sauces and condiments were highest in the 24 h MPR Traditional group (Fig. 4.7) and lowest in the FFQ Traditional dietary pattern (Fig. 4.6), a reason for this could be underestimated portion sizes in the FFQ. Overall, the foods consumed within the Traditional group from 24 h MPR data also corroborate a British traditional pattern defined in previous literature (Pryer et al., 2001).

The participants that scored highly on the Cosmopolitan dietary pattern in the FFQ (Fig. 4.6) also consumed considerably more legumes (229 g/day), vegetables (323 g/day), fish (92 g/day), tea and coffee (647 g/day) and slightly more wholegrains/meal, fats/oils and sweet desserts than the Traditional or Convenience groups in the 24 h MPR (Fig. 4.7; Appendix 9.16). As expected, this group consumed the lowest quantity of takeaway food (0 g/day), SSB (40 g/day), alcohol (36 g/day), meat (59 g/day), and sauces/condiments (9 g/day) (Appendix 9.16). Legumes and nuts scored negatively in the Cosmopolitan dietary pattern from the FFQ data (Fig. 4.6) and high in the 24 h MPR (Fig. 4.7), an explanation for this could be the underrepresentation of this food group in the FFQ design (Appendix 9.14) compared to the detailed nature of 24 h MPR dietary assessment. The characteristics from the Cosmopolitan group in the 24 h MPR were more suited to those labelled Mediterranean in previous research (Babio et al., 2009; del Mar Bibiloni et al., 2012); rich in fruits and vegetables, nuts, legumes, eggs, olive oil, and fish and seafood.

Finally, participants in the 24 h MPR Convenience group consumed more alcohol (292 g/day), SSB (473 g/day), refined grains (147 g/day), takeaways (80 g/day) and eggs (54 g/day) than the other groups (Fig. 4.7; Appendix 9.16). Lowest intakes were for wholegrains/meal (28 g/day), potatoes (35 g/day), fruit (91 g/day), fish (22 g/day), and fats/oils (22 g/day) (Appendix 9.16). Results verified the high scores for refined grains, fats and oils, eggs, takeaways from the FFQ Convenience dietary pattern (Table 4.7) and supported the findings from previous literature referring to convenience (Pryer et al., 2001) and Western dietary patterns (Hu et al., 2000; Wirfält et al., 2013; Englund-Ögge et al., 2014).

4.5.5 FFQ and 24 h MPR development

A significant role of the pilot was to develop and test the two dietary assessment techniques (FFQ and 24 h MPR). Findings from the pilot suggested that the FFQ was important for understanding the sample characteristics and gave a good general sense of habitual dietary intake. Whereas the 24 h MPR complemented the FFQ data, by adding fine detail to the foods consumed and allowed for validation. Feedback from participants suggested that the telephone 24 h MPR was comparable, if not favourable, to the face-to-face method due to less burden and time constraints; thus supporting the plan to use telephone 24 h MPR in the final study (Study 3). Likewise, in the EPIC study, Brustad et al. (2003) reported no significant differences between the data obtained through telephone 24 h dietary recalls when compared to face-to-face methods.

4.6 LIMITATIONS

The study limitations included the difficulty to extrapolate a small sample size, who were predominately white-British, university staff or students, who were educated to postgraduate level (Table 4.6). Therefore, caution is advised, as the results might not necessarily be representative of the UK population. Over and underestimating of common food groups with FFQ is well reported (Day et al., 2001; McKeown et al., 2001; Lietz et al., 2002), therefore this study validated results by including 24 h MPR data. Furthermore, dietary self-report data (FFQ and 24 h MPR) suffer from measurement error and objective measures of food intake i.e. biomarkers were not included. To address this limitation metabolite fingerprinting of biological markers was included in Study 3 (Chapter 6). When using PCA, it is recommended to use a larger number of participants than variables or a minimum of 150 participants (Pallant, 2013). Therefore, results might not be especially reliable for the sample size of 26 but the methods were successful in practice.

4.7 Conclusion

To conclude, the findings from the pilot study showed that PCA is an appropriate technique for dietary pattern analysis, generating three distinct dietary patterns; 'Traditional, 'Cosmopolitan' and 'Convenience'. This was an encouraging result owing to the small-scale of the study, which supports the notion that the larger proposed study (Study 2) may be able to define many dietary patterns including specific takeaway food dietary patterns. A particularly interesting finding was the elements of the diet that tracked together in the 'Convenience' dietary pattern; unhealthy habitual foods (refined grains, cakes, pastries etc.) and takeaway food. These findings suggested that takeaway food consumption was a marker of an unhealthy diet overall and could increase risks of obesity and non-communicable disease when consumed over a long period of time and in large quantities (Garcia et al., 2012; Donin et al., 2017). Variances in the types of takeaway food consumed may contribute to diverse dietary outcomes, thus further research to understand how opposing dietary patterns may affect diet and health is warranted, and to assess whether it is takeaway food or the remainder of the diet that influences poor dietary outcomes. Results from the 24 h MPR were also in agreement with the majority of characteristics in each dietary pattern, suggesting that the use of PCA on FFQ data is an acceptable measure of population dietary intakes (Nanri et al., 2012). After the successful development of suitable dietary assessment techniques and validation of the results, it was justified to carry out the subsequent studies 2 and 3 (Chapter 5 and 6 respectively). The collection of supplemental biomarker data (Lloyd et al., 2013) was also included into the project to add extra value to a pioneering project (Chapter 6), which has the potential to influence future research and interventions.

Chapter 5
Study 2

5 CHAPTER 5: TAKEAWAY FOOD HABITS AND WIDER DIETARY PATTERNS OF A MERSEYSIDE SAMPLE INCLUDING SOCIO-DEMOGRAPHIC AND LIFESTYLE CORRELATES

ABSTRACT

Background: Low-quality and nutrient-poor takeaway, take-out and fast foods have increased in popularity over the past 40 years and have been implicated as contributors to obesity and chronic disease. There has been much debate regarding the determinants of takeaway food consumption, however, it is evident that the cause is multi-faceted including societal, environmental, socio-economic, demographic, financial and behavioural factors. ***Aim of Study 2:*** Investigate takeaway food consumption and wider dietary patterns in a Merseyside sample and explore their socio-demographic and lifestyle determinants. ***Method:*** A cross-sectional observational study of 1724 adults (aged 18–64 years) was conducted in Merseyside, UK. Consumption of 212 habitual and takeaway foods respectively was measured using a food frequency questionnaire and socio-demographic and lifestyle factors were also self-reported. All data were analysed using SPSS; principal component analysis (PCA) determined dietary patterns and Spearman's correlation, Mann Whitney U and Kruskal Wallis H tested the relationship between takeaway food intake/dietary patterns with socio-demographic and lifestyle factors. ***Results:*** Being male, in a younger age group, having children in the household, alcohol use and smoking were positively associated ($p < 0.05$) with takeaway food consumption, whereas older age groups and higher qualifications were negatively ($p < 0.01$) associated. PCA identified four distinct dietary patterns; 'Western', 'Prudent', 'Takeaway' and 'Meat and Dairy', which were associated with a number of variables at varying degrees. ***Conclusion:*** The present study offers valuable insight into the behavioural and societal factors that may contribute to takeaway food consumption and wider dietary patterns in the UK adult population. These results could help inform policy and interventions to target the impacts of takeaway food on public health. Nevertheless, further understanding of the factors influencing consumption are required for the successful transition of a current unhealthy food environment to reduce its impact on obesity and related non-communicable disease.

5.1 CHAPTER OUTLINE

This was a cross-sectional (observational) study using the 'Eating Habits Survey' (EHS) dietary assessment method from the pilot study (Study 1, Chapter 4); however, a convenience sample of a Merseyside population was recruited as opposed to students and staff from LJMU. The EHS (the first layer of analysis) was designed to collect cross-sectional data on food consumption and socio-demographic and lifestyle correlates, thus creating a generalised overview of the dietary patterns and background of the study population. Self-reported habitual and takeaway food consumption, and self-reported demographic and anthropometric information were collected as mentioned previously (Chapter 4). Finally, data from the EHS was analysed and takeaway food consumer sub-groups were identified for the final study (Study 3, Chapter 6). This was to determine their dietary patterns from two forms of self-reported dietary assessment (food frequency questionnaire (FFQ) and 24 hour 'Multiple Pass' dietary recalls (24 h MPR)) and to complement a biomarker technique to classify food biomarkers in urine indicative of dietary exposure and takeaway foods.

5.2 Aims and objectives

Aim: To investigate takeaway food consumption within wider dietary patterns of the study population and their self-reported socio-demographic and lifestyle determinants.

Objectives:

1. Identify socio-demographic and lifestyle characteristics from a Merseyside sample.
2. Determine habitual and takeaway food consumption and associated socio-demographic, lifestyle and behavioural factors.
3. Classify dietary patterns of the participant sample and any associated determinants.

5.3 Methods

5.3.1 Study population and recruitment

The study sample (n = 1782) was recruited using advertisements (in social media, relevant websites, e-mails, media and locations in the community) and word of mouth from August 2016 to October 2017 (Figure 5.1). Participants received a link to a participant information sheet (Appendix 9.20) and provided implied consent when completing the online EHS. Convenience sampling (the most accessible participants were researched due to limited resources, time and workforce) (Etikan et al., 2016) and snowball sampling (participants were asked to refer friends or family to the survey to reach a wider “hard-to-reach” audience) (Atkinson and Flint, 2001; Heckathorn, 2011) were used in combination to meet the target sample size in a short time-frame. From the original sample, participants were removed who did not reside in Merseyside (n = 51) since a number of participants were from overseas and had a-typical characteristics and those from surrounding counties such as Cheshire have different demographic characteristics (more affluent, less takeaways, less CVD) than those in Merseyside. In addition, those who were extreme outliers for consumption of food groups (n = 7) were removed; for example when totalled, one participant reported consumption of 882.5 portions of vegetables/week and another participant reported 613 portions/week. Likewise, for refined grains, a participant reported consumption of 609 portions/week. These exclusions left a sample size of 1724.

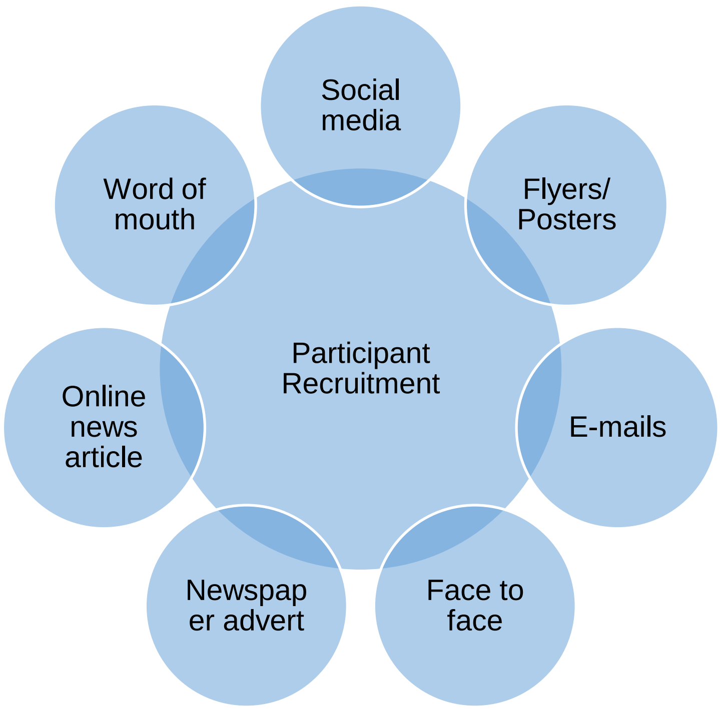


Figure 5-1 Recruitment techniques for Study 2

The predominant successful recruitment method was online via social media using ‘Facebook’, ‘Twitter’, and ‘Nextdoor’ campaigns to promote the study on a fortnightly basis (Appendix 9.21). ‘Facebook’ recruitment involved promotion on pages, sharing by others and publishing on multiple groups in Merseyside. ‘Twitter’ via the homepage and retweets and ‘Nextdoor’ (a local social network for people in the neighbourhood) by sharing invites to different areas pages in Merseyside. In addition, a newspaper advert was published in the ‘Liverpool Echo’ newspaper and online on Thursday 20th October 2016 (Appendix 9.22). LJMU campus displayed recruitment flyers/posters in locations including the canteen, hallways and on notice boards (Appendix 9.23) and promotion occurred in areas with takeaway food outlets, at community centres, churches, local shops, and youth centres in Merseyside. Students and staff across LJMU received direct e-mail invitations (from the researcher or member of the supervisory team) using university mailing lists. On two occasions, a study invitation in the LJMU library newsletter promoted the study. Finally, an online news article, published on ‘The Conversation’, endorsed the research and included a link to the study (see publications list on Page 23).

A power calculation estimated the initial target sample size for Study 2 (Table 5.1) (Naing et al., 2006; Charan and Biswas, 2013). It was estimated that there were 241,110 adult takeaway food consumers in Merseyside; 27% (Food Standards Agency, 2014) of 893,000 adults (439,800 males, 453,200 females) aged 16–64 years (Office for National Statistics, 2016). In the current study the confidence interval (z): 1.96, prevalence (p): 0.27 (27% takeaway consumers), and precision (d): 0.05. Therefore, 261.38 was the sample size needed in order to produce results that reflect the target population (takeaway food consumers) as precisely as needed (Table 5.1).

Table 5-1 Power calculation for takeaway food consumers in Merseyside

Equation	Calculation
$n = \frac{Z^2P(1-P)}{d^2}$ Where: *n* = sample size, *Z* = Z statistic for a level of confidence, *P* = expected prevalence or proportion (in proportion of one; if 20%, *P* = 0.2), and *d* = precision (in proportion of one; if 5%, *d* = 0.05).	$n = \frac{1.96^2 \times 0.27(1-0.27)}{0.05^2}$ $n = \frac{3.8416 \times 0.1701}{0.0025}$ n = 261.38

However, previous dietary pattern studies report larger sample sizes of between 700 and 5,000 and participation rates of between 37% and 98.5% (Mullie et al., 2010; Appannah et al., 2015; Funtikova et al., 2015; Granic et al., 2015) (Appendix 9.4). Moreover, the number of participants required for Study 3 (Chapter 6), a sub-sample of 200 from Study 2 set by IBERS, AU, needed to be considered. Therefore, taking note of previous literature, participant response and dropout rates, the sub-sample required for Study 3, and to maintain a realistic timeframe, a target of 2000 participants was estimated. The recruitment period was 14 months and thus 2000 participants was thought to be achievable.

5.3.2 Dietary assessment in Eating Habits Survey

Habitual diet and takeaway food consumption was characterised using the 'Eating Habits Survey' (EHS), designed according to the FFQ previously employed by the EPIC Study (Bingham et al., 1997; University of Cambridge, 2014), and had passed pilot testing (Chapter 4). Online Surveys (Jisc, UK) published an online version of the survey on 17th August 2016 (Appendix 9.24). Online only format was decided due to the positive feedback during pilot testing, the large target sample size (n = 2000) and to eliminate the chance of human error when entering data from hardcopy format into Microsoft Excel. Edits to the survey occurred on occasion during recruitment in response to participant feedback; initially participants commented on social media that they thought the further study (Study 3) was compulsory which deterred them from participating. Therefore, changes to the online EHS included moving information regarding the further study to the end of the survey (Appendix 9.24). After the opening of the EHS, geographical area of residence was included, thus participants who had not included this information and had consented to being contacted were e-mailed for postcode data. During the initial 4 months, recruitment was slower than anticipated, therefore a newspaper advert in the Liverpool Echo was organised in October 2016 and a question regarding evaluation of recruitment method was included in the survey (Appendix 9.24).

5.3.3 Covariates in Eating Habits Survey

The background information section contained in the EHS completed by all participants included self-reported questions on demographics, general health, anthropometrics and activity levels (Chapter 4). Demographics included gender, age group, ethnicity, marital status, living arrangements, number of adults and children in the household, education, employment status and occupation. Education was characterised as highest qualification gained ranging from no formal qualifications to postgraduate qualifications. Occupational class was condensed into 4 main groups based on the NS-SEC managerial/professional (including professional, managers, directors and senior officials), intermediate (including administrative and secretarial, sales and customer service), routine/manual (skilled trade, elementary, caring, leisure and other service, process, plant and machine), and student/unemployed/retired. A Likert scale measured self-reported health status (1 being very good and 5 being very poor) and a question inquired about any long-term illness that limited daily activity.

Physical activity level was assessed and included daily work (usually sitting, walk about quite a lot, usually lift and carry light loads, heavy work) and estimated number of hours per week in leisure time activities (mildly energetic, moderately energetic and vigorous), adapted from previous studies (Wareham et al., 2003; Fave et al., 2011). Self-reported anthropometric measurements were in preferred units (stone or Kg, cm, inches, or feet), and were converted into metric. Smoking habits were grouped as smoker (smoke daily, smoke occasionally) and non-smoker (former smoker or never smoked). A question inquired about the number of standard drinks of alcohol consumed per week or non-drinker status.

5.3.4 Statistical analysis

Data screening and cleaning was conducted using IBM SPSS® 24.0 (SPSS Inc., Chicago, Illinois, USA) to check for any outliers and errors on the categorical and continuous variables; seven outliers were removed from the data set as mentioned earlier (Methods 5.3.1). Descriptive statistics determined frequency of takeaway food consumption, takeaway food behaviours, socio-demographic, lifestyle, anthropometric and health variables. The Kolmogorov Smirnov test was used to explore the distribution of each variable and data was considered non-normal because the Sig. was $p < 0.05$. For example, takeaway food consumption was not evenly distributed as most individuals consumed 0–3 portions per week. Some extreme outliers were retained within the data because they were considered within acceptable range of consumption (e.g. takeaway food consumption of 4–5 times a day). These high scores skewed the data and affected the mean, meaning that the data were best represented by the median (Lumley et al., 2002). Therefore, non-parametric tests (Spearman's Rank Order Correlation (rho), Mann Whitney U and Kruskal Wallis H) explored correlations or differences between groups for takeaway food consumption and socio-demographics.

5.3.4.1 Principal component analysis to produce dietary patterns

The 212 food items from the EHS were grouped into 38 food groups (Table 5.2). Food groups were organised based on nutrient composition and with inspiration from a list of core and non-core foods described in the NDNS (Ziauddeen et al., 2017). A PCA (method as described in Chapter 4) derived dietary patterns from the data. However, after inspection of the correlation matrix it was decided to remove a number of food groups (red meat, fish, nuts and alcohol) that did not have correlations of $r = 0.3$ or greater, which left 34 food groups to re-test.

Table 5-2 Food grouping used in the dietary pattern analysis of the EHS cohort

Food group	Definition and content
1. Vegetables	Carrots, spinach, broccoli, brussel sprouts, cabbage, peas, green beans, courgettes, cauliflower, parsnips, leeks, onions, garlic, mushrooms, sweet peppers, beansprouts, mixed vegetables, vegetable soup
2. Salad	watercress, tomatoes, sweetcorn, beetroot, coleslaw, green salad,
3. Fruit	Apples, pears, oranges, grapefruit, bananas, grapes, melon, peaches, strawberries, avocado, tinned fruit, dried fruit
4. Nuts	All nuts
5. Legumes	Bakes beans, pulses, tofu,
6. Wholegrains/meal	Brown bread, wholemeal bread, porridge, all bran, wholegrain cereal, brown rice, wholemeal pasta
7. Refined grains	White bread, scones, crackers, pitta, garlic bread, sugary cereal, plain cereal, white rice, pasta, tinned pasta, noodles, Yorkshire puddings, lasagne
8. Buns/Cakes/Pastries	Cakes, buns, sweet pies, fruit pies
9. Biscuits	Chocolate biscuits, plain biscuits,
10. Fried potato	Crisps, chips, wedges, roast potatoes
11. Other Potato	Boiled potatoes, potato salad, baked potato
12. Chocolate	chocolate,
13. Sugar and sweets	sweets, sugar
14. Sweet condiments	Tomato sauces, ketchup, other sauces, relishes, marmite, dips, jam, peanut butter, chocolate spread
15. Red Meat	Beef, pork, lamb, liver, meat soup
16. Bacon and Ham	bacon, ham
17. Burgers/Processed Meat	beef burgers (not takeaway), processed meats, pies,
18. Sausages	sausages
19. Poultry	Chicken, turkey
20. Fish and shellfish	Fried fish, breaded fish, white fish, oily fish, shellfish
21. Milk (dairy)	dairy milk, coffee whitener,
22. Other milk (non-dairy)	milk from plant source (Soya, Almond, oat, rice)
23. Cheese	cheese, cottage cheese,
24. Yoghurt	full fat yoghurt, low fat yoghurt,
25. Dairy desserts	Single cream, double cream, dairy dessert, Milk puddings, ice cream,
26. Egg	Boiled, scrambled, fried, poached, quiche

27. Butter/Spreads/Oils	Butter, margarine, polyunsaturated margarine, other margarine, low fat spread, mayonnaise, low calorie mayonnaise, French dressing, salad dressing
28. Soups and savoury sauces	
29. Tea/Coffee/Diet drinks	Tea, Coffee, Low calorie SSB
30. Alcohol	Wine, beer, liquors, spirits
31. Sugar-sweetened beverages	Cocoa, Horlicks, SSB, pure fruit juice, cordial/squash
32. Pizza at home	Pizza (not takeaway)
33. Fast Food	Beef burger, chicken burger, fried chicken, fish burger, veggie burger, mozzarella sticks, fries, McDonald's breakfast, Subway, sauces and dips
34. Chinese	Chicken wings, spare ribs, crispy duck, spring rolls, prawn crackers, siu mai, skewered chicken, soup, salt and pepper chicken, peking dish, lemon/orange/plum sauce dish, satay dish, sweet and sour, curry, chop suey, kung po, foo yung, chow mein, beef dish, chicken or Duck Veg, king prawn, fried rice, salt and pepper chips, noodles, boiled rice, chinese sauces
35. Indian	Poppadums, samosa, tikka starter, tandoori, korma, tikka masala, rogan josh, bhuna, biryani, balti, sagwala, tikka jalfraizi, dupiaza, medium curry, achari, vindaloo, pathia, pasanda, special, butter chicken, saag, dal, naan, paratha, pilau, fried rice, boiled rice
36. English	Fish, scampi, sausage, omelette, roast chicken, chips, sauces
37. Pizza	Cheese, meat, seafood, vegetarian, garlic bread
38. Kebab	Donner, chicken, sheesh

NB: Food groups removed from final PCA analysis highlighted in red

An orthogonal (varimax) rotation (Thurstone, 1947) was performed on the 34 food groups to aid interpretability. This technique redistributes the explained variance for the individual components and so simplifying the factor structure and increasing the number of larger and smaller loadings (Northstone et al., 2008; Field, 2013; Roberts et al., 2018). Once the components were produced it was necessary to decide on how many components to retain by using four major criteria; 1) the eigenvalue-one criterion, (2) the proportion of total variance accounted for, (3) the scree plot test, and (4) the interpretability criterion.

Four principal components were retained, based on components that had an eigenvalue > 1.0 and a graphical evaluation of the scree plot of eigenvalues (i.e., the point at which the slope of the plot changes), in accordance to previous research (Fransen et al., 2014). Other studies have used absolute factor loading > 0.20 to define food groups making meaningful contribution to dietary patterns (Slattery, 2010; Smith et al., 2013; Atkins et al., 2016). However, from statistical guidance it was decided to only include component scores > 0.30 (Pallant, 2013). To retain four components from the 34 a forced Factor Extraction was run to produce results.

5.3.4.2 Socio-demographic and lifestyle correlates or differences between groups

The component scores for each participant for each of the four dietary patterns were used to plot against socio-demographic, lifestyle and anthropometric variables. Data was considered non-normal (as assessed by Kolmogorov Smirnov) and the median was considered as best representation of the data. Therefore Spearman's rho was used as the non-parametric alternate to Pearson's correlation to determine if any of the four dietary patterns were associated with the socio-demographic variables. Mann-Whitney U was used to show difference between groups of variables with two categories (gender, ethnicity) and Kruskla-Wallis H was run to show difference between groups of variables with more than 2 categories (occupation, marital status).

5.4 RESULTS

5.4.1 PARTICIPANT CHARACTERISTICS

The total sample was 1724 (1424 female, 297 male and 3 transgender) participants from Merseyside (Table 5.3). Fifty-one exclusions were made due to participants residing outside of the study area and a further seven for being extreme outliers concerning servings of foods consumed (described earlier). All data was recorded for dietary intake; however, there was some missing data for height and weight and waist circumference (Table 5.4).

Table 5-3 Study population socio-demographic characteristics 2016–2017 (n = 1724)

Variable	Level	*n*	%
Age group (years)	18 – 25	397	23.0
	26 – 35	516	29.9
	36 – 45	382	22.2
	46 – 55	255	14.8
	56 – 64	174	10.1
Gender	Female	1424	82.6
	Male	297	17.2
	Transgender	3	0.2
Ethnicity	White	1655	96.0
	Non-White	69	4.0
Marital status	Single	865	50.2
	Married (including civil partnership)	717	41.6
	Divorced/Separated/Widowed	142	8.2
Living arrangements	On own	216	12.5
	Wife/Husband/Partner	913	53.0
	Parents	184	10.7
	Friends	92	5.3

	Children	275	16.0
	Other	44	2.6
Adults in household (18+)	1	375	21.8
	2	913	53.0
	3	251	14.6
	4 or more	185	10.7
Children in household (U18)	0	906	52.6
	1	373	21.6
	2	317	18.4
	3	91	5.3
	4 or more	37	2.1
Educational achievement	No formal qualifications	67	3.9
	GCSE/O-Level	290	16.8
	A-Level or Equivalent	487	28.2
	Degree Level	503	29.2
	Postgraduate Qualification	306	17.7
	Other	71	4.1
Employment status	Employed/self-employed	1099	63.7
	Not in employment	186	10.8
	Full Time Student	262	15.2
	Retired	83	4.8
	Other (carer, ill health)	94	5.5
NS-SEC	Managerial/Professional	547	31.7
	Intermediate	347	20.1
	Routine/Manual	400	23.2
	Student/Unemployed/Retired	430	24.9

NB: *n* = number of participants. NS-SEC: National Statistics Socio-economic Classifications. U18 = Under 18 years of age.

Table 5-4 Study population lifestyle and self-reported anthropometric characteristics 2016-2017 (n = 1724)

Variable	Level	*n*	%	Median (IQR)
BMI (kg/m²)		1693		26.5 (23.2-–31.3)
WHtR		1000		0.48 (0.5–0.5)
Alcohol consumption (drinks/week)				1.5 (0.0–4.5)
	0	609	35.3	
	0.1–4	470	27.3	
	4.1–13.9	465	27.0	
	> 14	180	10.4	
Daily leisure activity (minutes)	LPA	1724		60 (34.3–120.0)
	MVPA	1724		34.3 (17.1–68.6)
Smoking status	Smoker	397	23.0	
	Non-Smoker	1327	77.0	
Daily occupational activity	Sedentary	716	41.5	
	Standing	777	45.1	
	Manual	204	11.8	
	Heavy	27	1.6	
Supplement use	Yes	714	41.4	
	No	975	56.6	
	Not Sure	35	2.0	
Health status	Very Good	293	17.0	
	Good	584	33.9	
	Neither Good nor Poor	608	35.3	
	Poor	191	11.1	
	Very Poor	48	2.8	
Long-term illness	Yes	502	29.1	
	No	1222	70.9	

NB: n = number of participants. IQR = Inner Quartile Range. LPA = Low physical activity. MVPA = Moderate and vigorous physical activity.

5.4.2 Frequency of takeaway food consumption

Self-reported total frequency of takeaway food consumption showed that the majority (46%) of participants (n = 1724) consumed takeaway food 1–3 times per month. This was followed by 26% of participants consuming takeaway food once a week, 18% never or less than once per month, 9% 2–4 times per week and 1% consuming takeaway food 5-6 times a week or more, respectively (Fig. 5.2).

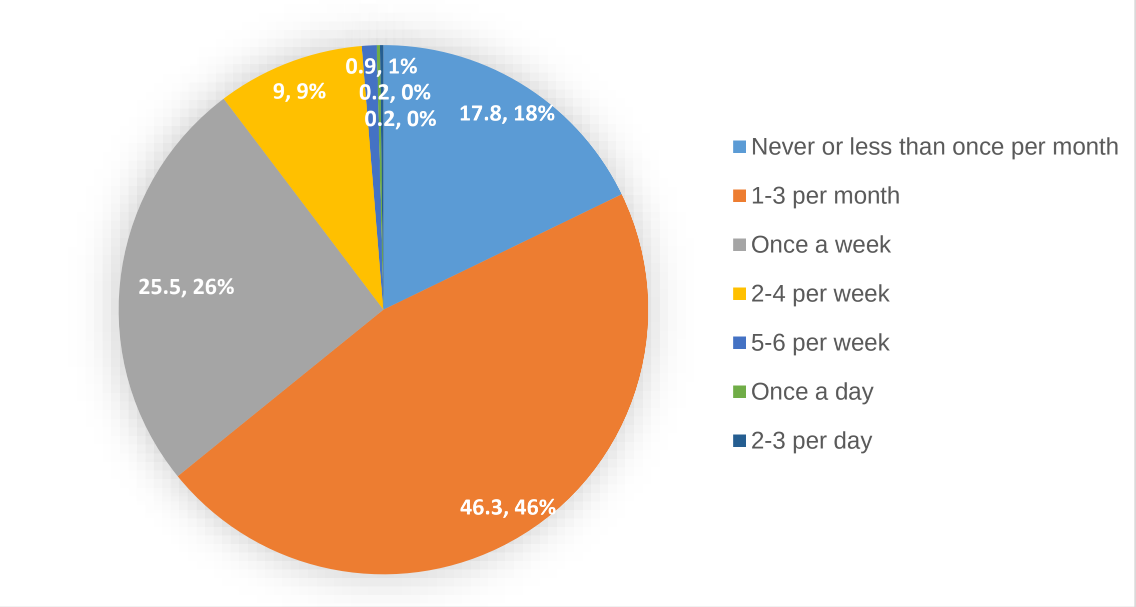


Figure 5-2 Frequency of takeaway food consumption (single question) (n = 1724)

When calculating the total frequency of takeaway food consumption from the EHS aggregated food groups, 29% of participants reported consuming 2–3 servings of takeaway food per day. This was followed by 21% consuming 1 serving per day, 16% 2–4 servings per week, 12% 5–6 servings per week, 6% never or less than once per month, 5% 4–5 times per day, 4% once a week, 4% 6+ times per day and 3% 1–3 times per month, respectively (Fig. 5.3). Frequency of takeaway food consumption from the aggregated EHS food groups (Fig. 5.3) was higher than self-reported frequency (Fig. 5.2), which suggests that the aggregation of a large number of items into a variable may have overestimated consumption, a commonly reported limitation of FFQs (Smith et al., 2009).

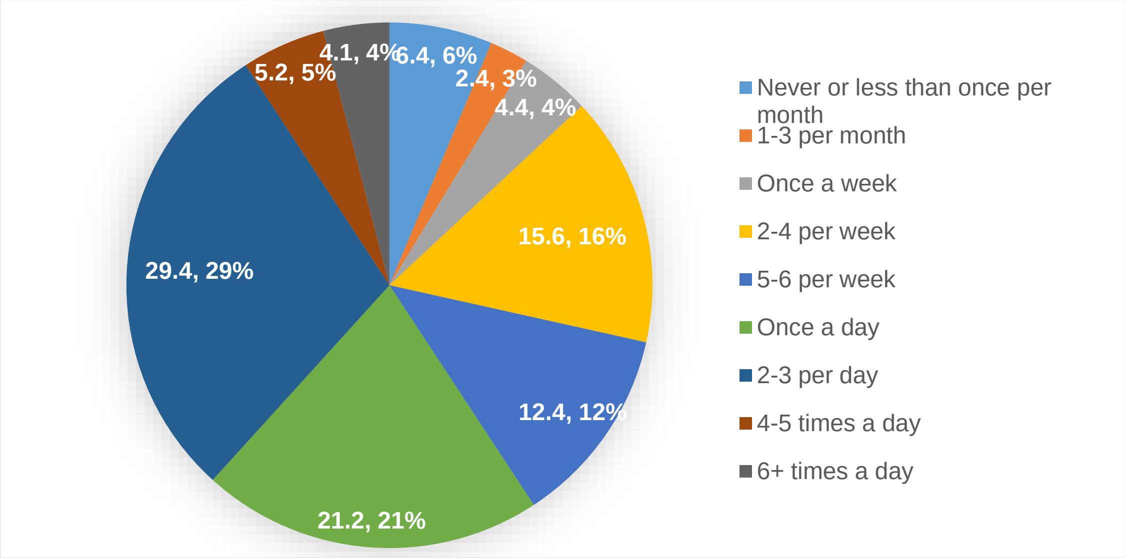


Figure 5-3 Frequency of takeaway food consumption (aggregated EHS data) (n = 1724)

Chinese was the most commonly consumed takeaway food, closely followed by American fast food, Indian, English, pizza and kebab. Other takeaway foods commonly consumed by the participants included Japanese (sushi), Thai, Greek, Lebanese, Mexican, Nando's. rotisserie chicken, Subway salad and KFC (Fig. 5.4).

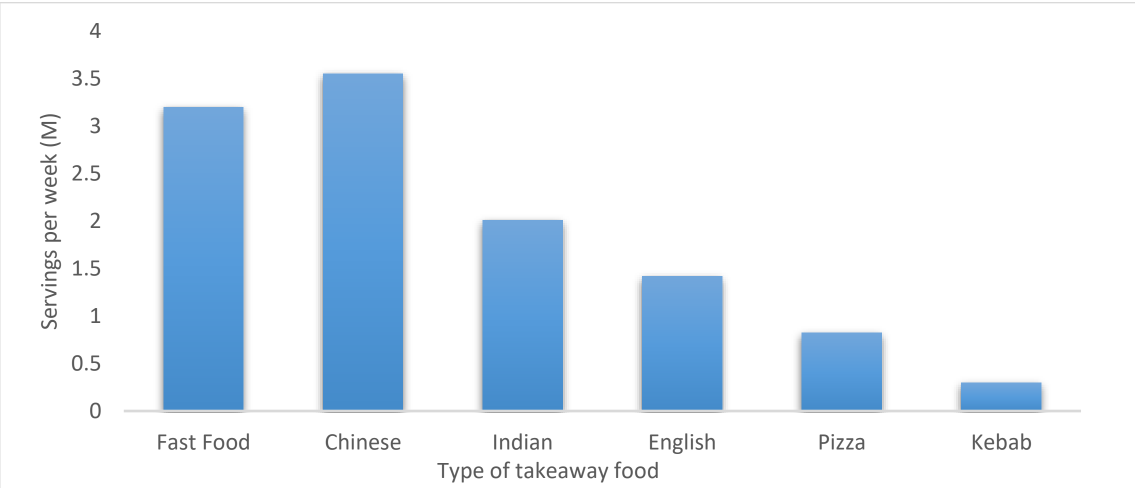


Figure 5-4 What type of takeaway/fast food do you eat most often? (Please tick all that apply) (n = 1724)

5.4.3 SOCIO-DEMOGRAPHIC DETERMINANTS OF TAKEAWAY FOOD CONSUMPTION

5.4.3.1 SPEARMAN'S CORRELATIONS

The relationship between takeaway food consumption and socio-demographic and lifestyle variables were investigated using Spearman's correlation. There were a number of statistically significant positive and negative correlations (Table 5.5). There was a significant ($p < 0.005$) small positive correlation between total takeaway food consumption and health status ($r = 0.24$), smoking frequency ($r = 0.11$), having children in the household ($r = 0.09$), moderate and vigorous physical activity ($r = 0.09$), daily occupational activity ($r = 0.09$), BMI ($r = 0.08$), alcohol consumption ($r = 0.07$), and adults in the household ($r = 0.06$). There was a significant ($p < 0.0005$) small negative correlation between total takeaway food consumption and age ($r = -0.27$), with older individuals consuming less takeaway food, and educational level ($r = -0.16$) with more educated participants consuming less takeaways.

Table 5-5 Spearman's rho correlations for total takeaway food consumption and socio-demographic characteristics and lifestyle factors

Socio-demographic/lifestyle factors	R	P value	n
Age Group	−0.27	< 0.0005	1724
Adults in household	0.06	0.010	1724
Children in household	0.09	< 0.0005	1724
Education level	−0.16	< 0.0005	1724
Health Status (Very good to Very poor)	0.24	< 0.0005	1724
Smoking frequency	0.11	< 0.0005	1724
Alcohol (drinks/week)	0.07	0.005	1720
Body Mass Index	0.08	0.001	1693

WHtR	−0.14	0.664	1000
Daily occupational activity	0.09	< 0.0005	1724
Low physical activity (min/day)	0.03	0.208	1724
Moderate and vigorous physical activity (min/day)	0.09	< 0.0005	1724

NB: R = Correlation coefficient. P Value = statistical sig. < 0.05. n = number of participants.

Spearman's rho investigated the relationship between each takeaway food group consumed (fast food, Chinese, Indian, English, pizza and kebab) and socio-demographic and lifestyle variables (Table 5.6). Increased age group was significantly ($p < 0.005$) moderately negatively correlated with consumption of fast food ($r = -0.40$) and pizza ($r = -0.34$) (Table 5.6; Fig. 5.5). There was a significant small negative correlation between age group and Chinese ($r = -0.15$), English ($r = -0.10$) and kebab ($r = -0.18$), and no correlation between Indian and age group.

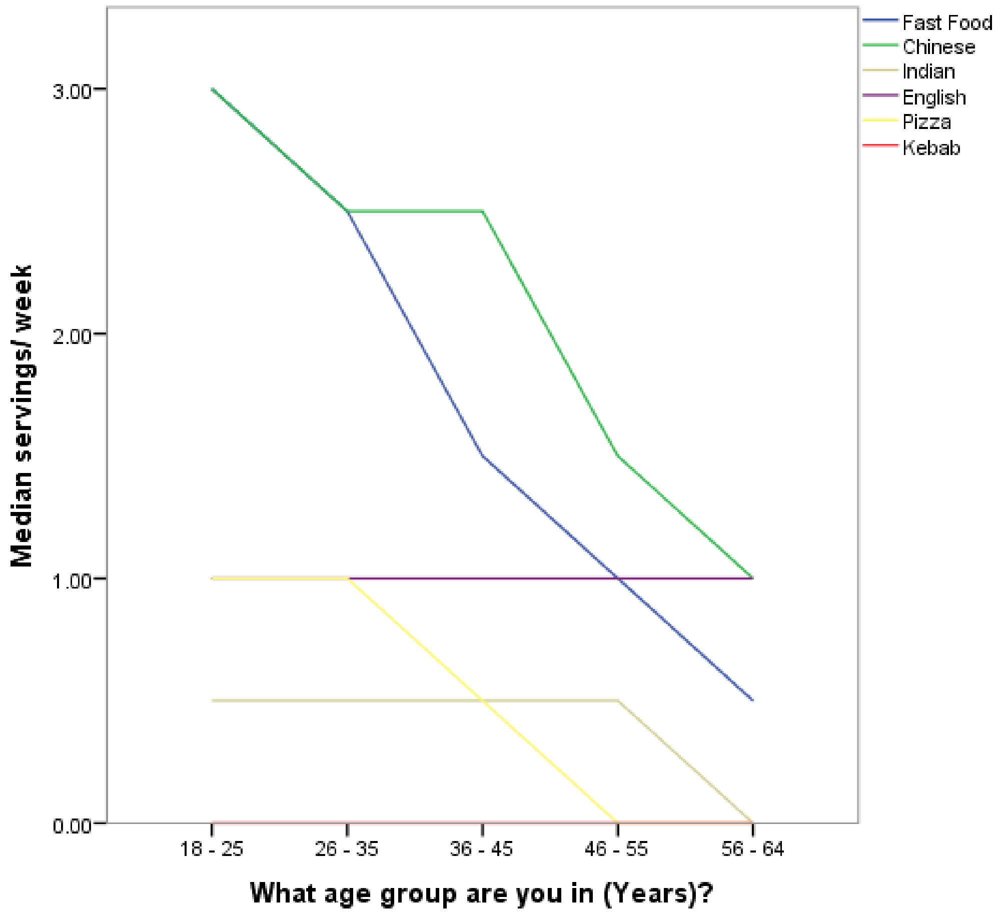


Figure 5-5 Correlations between six takeaway food categories and age group

There was a small positive correlation between having adults in the household and fast food (r = 0.05) and Chinese (r = 0.06) and no correlations with other takeaway food types (Table 5.6). Having children in the household was significantly ($p < 0.005$) correlated with consumption of fast food (r = 0.10), Chinese (r = 0.09), English (r = 0.07) and kebab (r = 0.07) and there was no correlation with Indian or pizza (Table 5.6). There was a significant ($p < 0.0005$) small negative correlation between educational level and fast food (r = −0.16), Chinese (r = −0.15), English (r = −0.18) pizza (r = −0.09) and kebab (r = −0.14) with more educated participants consuming fewer takeaways (Table 5.6; Fig. 5.6). There was no correlation between education level and Indian takeaway (r = 0.02).

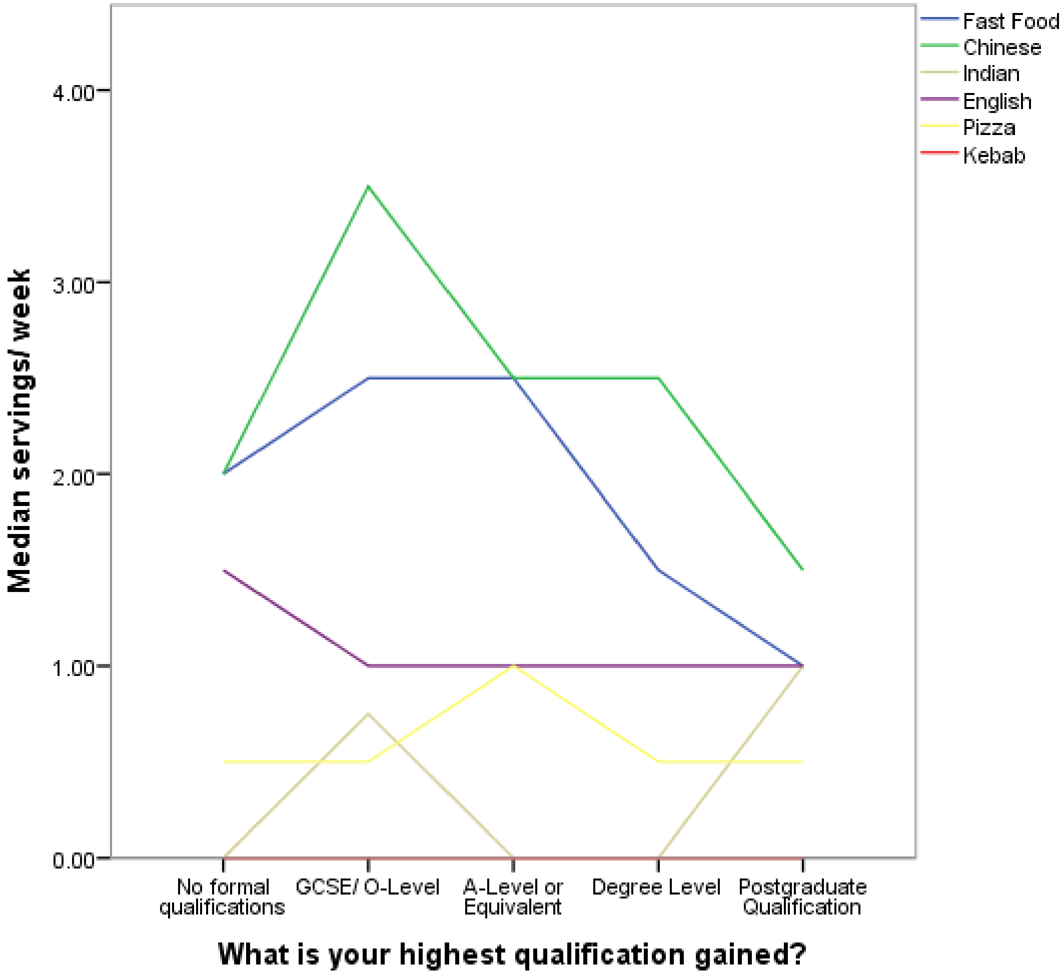


Figure 5-6 Correlations between six takeaway food categories and education

Smoking was significantly ($p < 0.005$) positively correlated with fast food ($r = 0.15$), Chinese ($r = 0.07$), English ($r = 0.09$), pizza ($r = 0.09$) and kebab ($r = 0.13$) however, there was no correlation with Indian takeaway ($r = -0.01$) (Table 5.6). There was a small positive correlation between alcohol consumption and Chinese (0.07) and Indian (0.16) but none so for fast food, English, pizza or kebab (Table 5.6). There was a significant ($p < 0.005$) small positive correlation with health status and fast food ($r = 0.26$), Chinese ($r = 0.18$), English ($r = 0.18$), pizza ($r = 0.17$) and kebab ($r = 0.13$) (Table 5.6; Fig. 5.7).

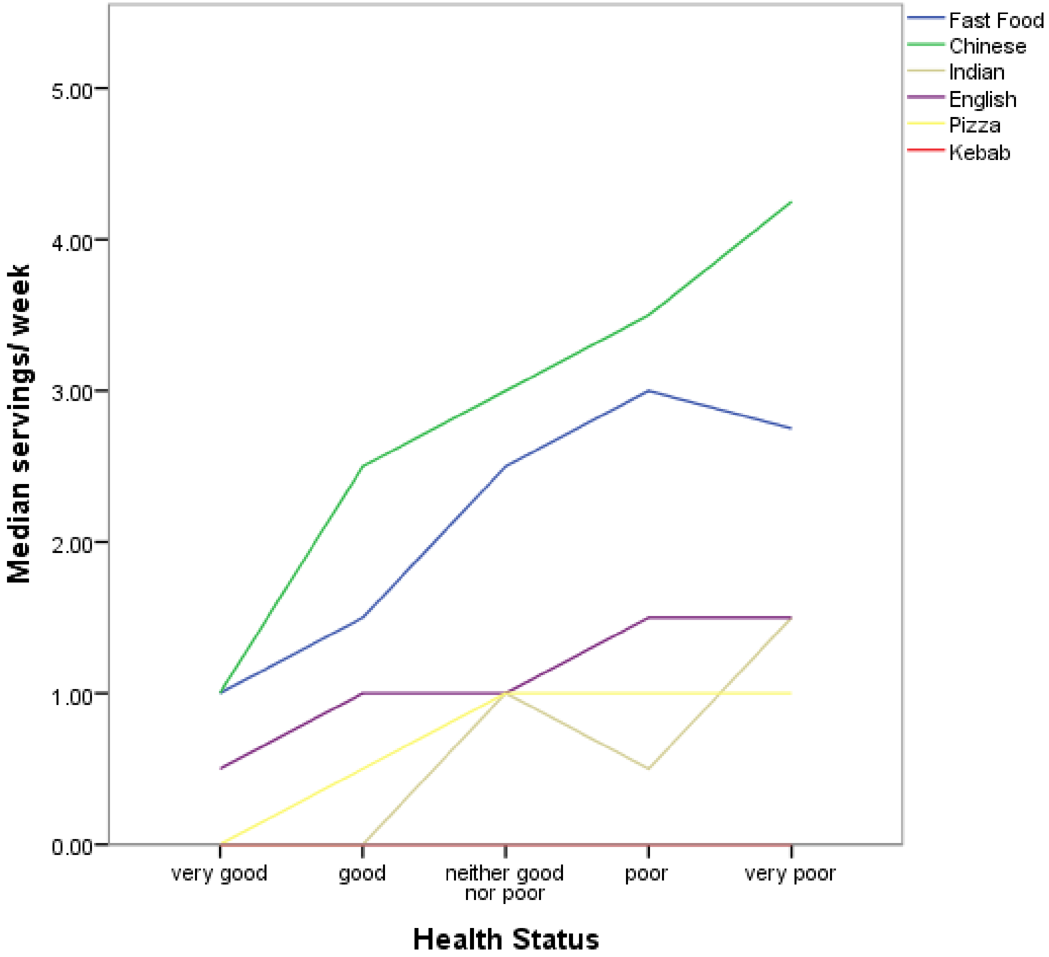


Figure 5-7 Correlations between six takeaway food categories and health status

BMI had a significant small positive correlation with fast food consumption ($r = 0.10$), Chinese ($r = 0.08$) English ($r = 0.08$) and kebab ($r = 0.12$), and no correlation with Indian ($r = -0.02$) or pizza ($r = 0.01$) (Table 5.6). There were no correlations between WHtR and any of the takeaway food categories (Table 5.6). Daily occupational activity had a significant ($p < 0.05$) small positive correlation with fast food ($r = 0.10$), Chinese ($r = 0.07$), English ($r = 0.09$) and pizza ($r = 0.06$) and no correlation with Indian ($r = 0.02$) or kebab ($r = 0.02$) (Table 5.6; Fig. 5.8).

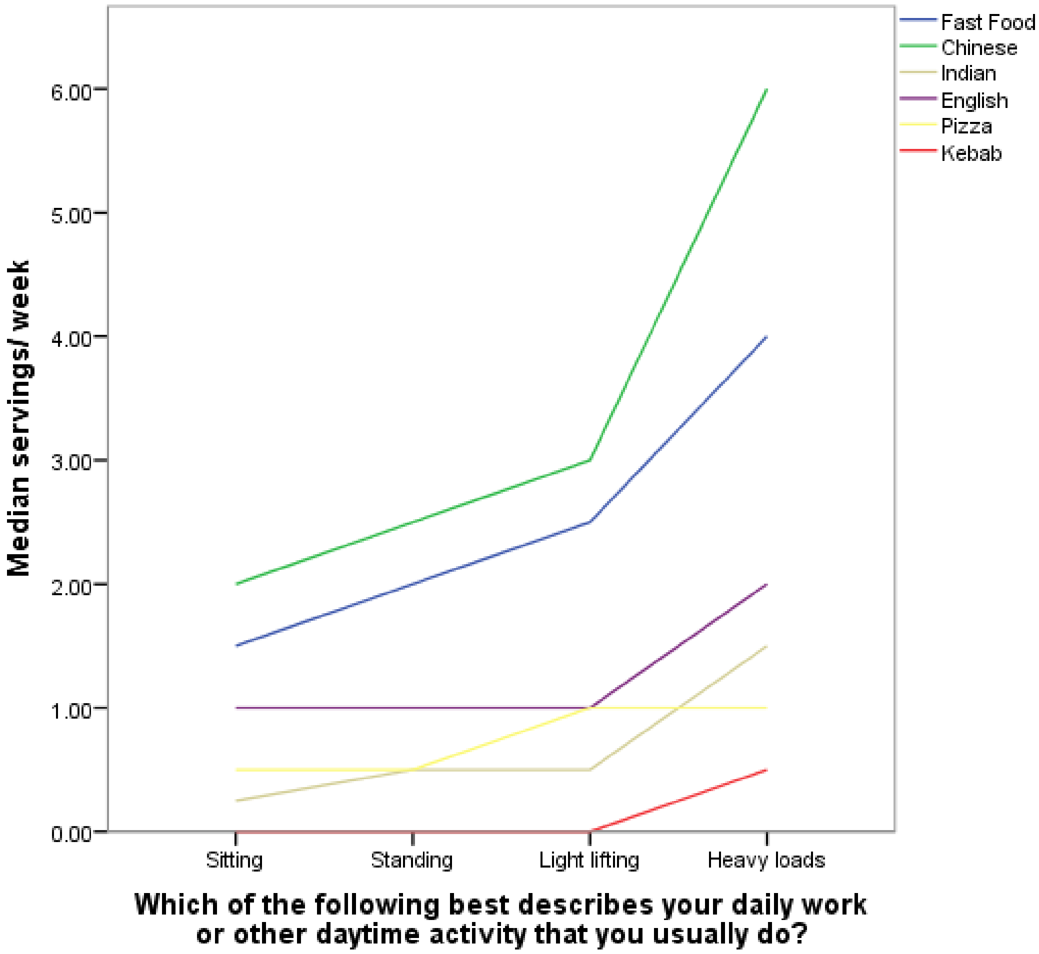


Figure 5-8 Correlations between six takeaway food categories and daily work activity

There was a significant ($p < 0.05$) small positive correlation between low physical activity and fast food ($r = 0.06$) and no correlation with any of the other takeaway food categories (Table 5.6). Moderate and vigorous physical activity was significantly ($p < 0.05$) positively correlated with fast food ($r = 0.10$), Chinese ($r = 0.06$), pizza ($r = 0.08$) and kebab ($r = 0.06$), with no correlations for Indian ($r = 0.04$) or English ($r = 0.03$) takeaways (Table 5.6).

Table 5-6 Spearman's correlation for dietary patterns and socio-demographic and lifestyle variables

Variables	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
1. Fast food	1																	
2. Chinese	.48**	1																
3. Indian	.20**	.38**	1															
4. English	.48**	.38**	.21**	1														
5. Pizza	.50**	.32**	.22**	.39**	1													
6. Kebab	.38**	.35**	.21**	.29**	.31**	1												
7. Age group	-.40**	-.15**	-.01	-.10**	-.34**	-.18**	1											
8. Adults	.05*	.06*	.04	.03	.04	-.01	-.17**	1										
9. Children	.10**	.09**	.01	.07**	.02	.07**	.03	-.10**	1									
10. Education	-.16**	-.15**	.02	-.18**	-.09**	-.14**	-.02	-.04	-.11**	1								
11. Health status	.26**	.18**	.05	.18**	.17**	.13**	-11**	.00	.05*	-.12**	1							
12. Smoking frequency	.15**	.07**	-.01	.09**	.09**	.13**	-.06*	.08**	.00	-.15**	.09*	1						

13. Alcohol frequency	-.00	.07**	.16**	-.02	.01	.01	.05*	.05	-.13**	.07*	-.08	.06	1					
14. BMI	.10**	.08**	-.02	.08**	.01	.12**	.16**	-.04	.08**	-.13	.34**	-.05*	-.11*	1				
15. WHtR	.01	-.03	-.04	.01	.04	-.03	-.01	-.02	-.02	.01	.02	-.00	-.02	-.01	1			
16. Daily work activity	.10**	.07**	.02	.09**	.06*	.02	-.11	.00	.10**	-.12**	-.09**	.11**	-.00	-.08**	-.05	1		
17. LPA	.06*	.02	-.02	-.01	.02	-.02	-.14**	.02	.07**	-.04	-.08	.04	.03	-.05*	.00	.28**	1	
18. MVPA	.10**	.06*	.04	.03	.08**	.06*	-.14**	.02	.04	-.04	-.19**	.03	.02	-.18**	. -.01	26**	.41**	1

NB: **Correlation is significant at the 0.01 level (2-tailed). *Correlation is significant at the 0.05 level (2-tailed). BMI = Body Mass Index. WHtR = Waist to height ratio. LPA = Low physical activity, MVPA = Moderate and vigorous physical activity.

5.4.3.2 MANN-WHITNEY U TEST

A Mann-Whitney U test determined if there were differences in takeaway food consumption between dichotomous variables (groups of variables with two categories) such as gender (Table 5.7). Distributions of total takeaway food consumption for males and females were similar, as assessed by visual inspection (Pallant, 2013). Median total takeaway food consumption was statistically ($p = 0.001$) significantly higher in males (MED 9.5, IQR 4.5–17.5) than in females (MED 8.0, IQR 4.0–13.5) (Table 5.7). For ethnicity, distributions of the takeaway food consumption for whites and non-whites were not similar and mean ranks for takeaway food consumption was not statistically ($p = 0.105$) different between whites and non-whites. For supplement users, distributions were similar and median consumption of total takeaway food was significantly ($p < 0.0005$) higher in non-supplement users (MED 9.0, IQR 4.5–15.0) than in supplement users (MED 7.0, IQR 3.5–12.5) (Table 5.7). Takeaway food consumption was not significantly different for participants suffering with long-term illness compared to those not suffering a long-term illness.

Table 5-7 Mann Whitney U tests for total takeaway food consumption and background variables

	Similar distribution	U	Z	P value
Gender	Yes	185,938	−3.278	0.001
Ethnicity	No	63,668	1.622	0.105
Supplements	Yes	366,495	2.274	< 0.0005
Illness	Yes	319,566	1.368	0.171

NB: U = Mann Whitney U. Z = Standardised test statistic. P Value = statistical sig. < 0.05.

A Mann-Whitney U test determined if there were differences in each takeaway food category consumed with the same dichotomous variables (Table 5.8). Males consumed significantly ($p < 0.05$) more fast food, English takeaway, pizza and kebab than females. For ethnicity, non-whites consumed significantly ($p < 0.05$) more fast food and pizza than their white counterparts. Median consumption of fast food, Chinese, English, pizza and kebab was significantly ($p < 0.05$) higher in non-supplement users than in supplement users. Participants with no long-term illness consumed significantly ($p < 0.01$) more Indian and pizza takeaway than those suffering long-term illness.

Table 5-8 Mann Whitney U tests for differences between takeaway food categories and socio-demographic or lifestyle variables

Variables	Takeaway food categories						
	Fast Food	Chinese	Indian	English	Pizza	Kebab	Total TF
Gender (n = 1721)							
Male	2.5 (0.5–4.5)	2.5 (0.0–5.5)	1.0 (0.0–3.5)	1.0 (0.0–2.0)	0.5 (0.0–1.5)	0.0 (0.0–0.5)	9.5 (4.5–17.5)
Female	2.0 (0.5–3.5)	2.5 (0.0–4.5)	0.5 (0.0–3.0)	1.0 (0.0–1.5)	0.5 (0.0–1.0)	0.0 (0.0–0.5)	8.0 (4.0–13.5)
p-value	0.003	0.849	0.120	0.014	0.024	< 0.0005	0.001
Ethnicity (n = 1724)							
White	2.0 (0.5–3.5)	2.5 (0.0–5.0)	0.5 (0.0–3.0)	1.0 (0.0–1.5)	0.5 (0.0–1.0)	0.0 (0.0–0.5)	8.5 (4.0–14.0)
Non-white	2.5 (1.0–5.5)	1.5 (0.0–5.5)	0.5 (0.0–4.8)	1.0 (0.0–1.5)	1.0 (0.0–2.0)	0.0 (0.0–1.0)	9.5 (4.0–24.3)
p-value	0.030	0.293	0.130	0.726	0.015	0.093	0.105
Supplements (n = 1689)							
Yes	1.5 (0.5–3.5)	2.0 (0.0–4.5)	0.5 (0.0–3.0)	1.0 (0.0–1.0)	0.5 (0.0–1.0)	0.0 (0.0–0.5)	7.0 (3.5–12.5)
No	2.0 (0.5–4.0)	2.5 (0.0–5.0)	0.5 (0.0–3.0)	1.0 (0.0–1.0)	0.5 (0.0–1.0)	0.0 (0.0–0.5)	9.0 (4.5–15.0)
p-value	< 0.0005	0.004	0.593	0.003	0.002	0.023	< 0.0005
Illness (n =1724)							
Yes	1.5 (0.5–3.5)	2.5 (0.5–4.5)	0.0 (0.0–2.5)	1.0 (0.0–2.0)	0.5 (0.0–1.0)	0.0 (0.0–0.5)	8.0 (4.0–12.5)
No	2.0 (0.5–4.0)	2.5 (0.0–5.0)	0.5 (0.0–3.0)	1.0 (0.0–1.5)	0.5 (0.0–1.0)	0.0 (0.0–0.5)	8.5 (4.0–14.5)
p-value	0.385	0.908	0.002	0.103	0.005	0.932	0.171

NB: All values are median intakes unless otherwise stated. Interquartile range (25%–75%). n = number of participants.

5.4.3.3 KRUSKAL-WALLIS H TEST

A Kruskal-Wallis H test (sometimes also called the "one-way ANOVA on ranks") showed the difference between groups of variables with more than two categories for takeaway consumption (Table 5.9). Distributions for all variables were similar as assessed by visual inspection therefore; median intakes were included for comparison. For marital status, single participants consumed significantly ($p < 0.0005$) more fast food and pizza than married or divorced participants. Whereas, married participants consumed significantly ($p < 0.05$) more Indian takeaway food than single or divorced participants. Results on employment status showed that retired participants consumed significantly ($p < 0.0005$) less fast food, Chinese, Indian, English, pizza and total takeaway food than those in employment, students, unemployed or other. Total takeaway food consumption, specifically fast food, Chinese, English and Pizza was significantly ($p < 0.0005$) higher for participants in a manual occupation than for those in intermediate or managerial professions or for the unemployed. There was no significant difference for Indian takeaway food consumption between all NS-SEC.

Table 5-9 Kruskal-Wallis H tests for takeaway food categories and background variables (n = 1724)

Variables	Takeaway food categories						
	Fast Food	Chinese	Indian	English	Pizza	Kebab	Total takeaway
Marital status							
Single	2.5 (1.0–4.5)	2.5 (0.0–5.0)	0.0 (0.0–3.0)	1.0 (0.0–2.0)	1.0 (0.0–1.5)	0.0 (0.0–0.5)	9.0 (4.5–15.5)
Married	1.5 (0.0–3.0)	2.0 (0.0–4.5)	1.0 (0.0–3.25)	1.0 (0.0–1.5)	0.5 (0.0–1.0)	0.0 (0.0–0.5)	7.5 (4.0–12.5)
Divorced	1.0 (0.0–2.5)	2.25 (0.0–4.5)	0.0 (0.0–3.0)	1.0 (0.0–1.5)	0.0 (0.0–1.0)	0.0 (0.0–0.0)	6.0 (3.0–11.5)
p-value	< 0.0005	0.087	0.033	0.001	< 0.0005	0.001	< 0.0005
Employment status							
Employed	2.0 (0.5–3.5)	2.5 (0.0–5.0)	0.5 (0.0–3.0)	1.0 (0.0–1.5)	0.5 (0.0–1.0)	0.0 (0.0–0.5)	8.5 (4.0–14.0)
Unemployed	2.5 (1.0–4.0)	2.0 (0.5–5.0)	0.5 (0.0–2.5)	1.0 (0.5–2.0)	0.5 (0.0–1.5)	0.0 (0.0–0.5)	9.0 (5.0–16.0)
Student	2.5 (1.0–4.6)	2.5 (0.0–4.5)	0.0 (0.0–3.0)	1.0 (0.0–1.5)	1.0 (0.0–1.5)	0.0 (0.0–0.5)	8.75 (4.5–14.0)
Retired	0.0 (0.0–1.0)	0.5 (0.0–3.0)	0.0 (0.0–1.5)	0.5 (0.0–0.5)	0.0 (0.0–0.0)	0.0 (0.0–0.0)	3.0 (0.5–6.5)
Other*	2.0 (0.5–4.0)	3.0 (1.0–6.0)	1.0 (0.0–3.0)	1.5 (1.0–2.0)	0.5 (0.0–1.0)	0.0 (0.0–0.5)	9.0 (5.5–14.5)
p-value	< 0.0005	< 0.0005	0.014	< 0.0005	< 0.0005	< 0.0005	< 0.0005
NS-SEC							
Managerial	1.5 (0.0–3.0)	2.0 (0.0–4.5)	0.5 (0.0–3.0)	1.0 (0.0–1.5)	0.5 (0.0–1.0)	0.0 (0.0–0.5	7.5 (3.0–12.5)
Intermediate	1.5 (0.0–3.5)	2.0 (0.0–5.0)	0.5 (0.0–3.0)	1.0 (0.0–1.5)	0.5 (0.0–1.0)	0.0 (0.0–0.5	8.0 (4.0–13.0)
Manual	2.5 (1.0–4.5)	3.0 (1.0–5.5)	0.5 (0.0–3.0)	1.5 (0.6–2.0)	1.0 (0.0–1.5)	0.0 (0.0–0.5	10.0 (5.5–15.5)
Unemployed	2.0 (1.0–4.0)	2.0 (0.0–4.6)	0.0 (0.0–2.6)	1.0 (0.0–2.0)	0.5 (0.0–1.5)	0.0 (0.0–0.5	8.0 (4.0–14.5)
p-value	< 0.0005	< 0.0005	0.282	< 0.0005	< 0.0005	< 0.0005	< 0.0005

NB: All values are median intakes unless otherwise stated. Interquartile range (25%–75%). *Other = ill health, disability, carer or maternity leave. NS-SEC: National Statistics Socio-economic Classifications.

5.4.4 TAKEAWAY FOOD CONSUMER BEHAVIOUR

The motives for takeaway food consumption in the study sample included ‘as a treat’ (n = 1203), ‘they are easily available’ (n = 732), ‘I like the taste’ (n = 552) and ‘for social occasions’ (n = 510) (Fig. 5.9). Other main reasons were that ‘they are available close to my home’ (n = 428), ‘my friends and family like it’ (n = 357), and ‘I am usually too busy to cook’ (n = 410). Some other reasons for takeaway food consumption described by participants included ‘for convenience’ (n = 15), ‘to have a break from cooking’ (n = 12), ‘due to health issues or disability’ (n = 9), ‘being hungover or after a night out’ (n = 9) and ‘being too tired to cook’ (n = 8).

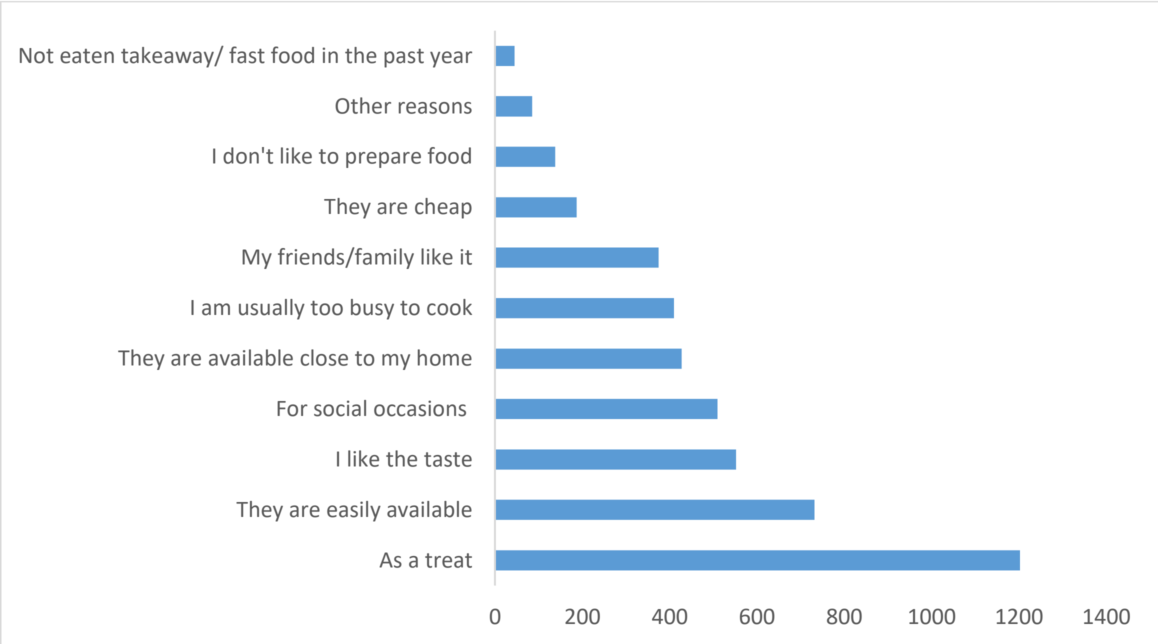


Figure 5-9 Why do you eat takeaway/fast food? (Please tick all that apply) (n = 1724)

The majority of the sample consumed takeaway food at home (n = 1589), followed by at friends/families (n = 548) and at the takeaway food outlet (n = 286). Other places included in the car, at work, camping and hotels (Fig. 5.10).

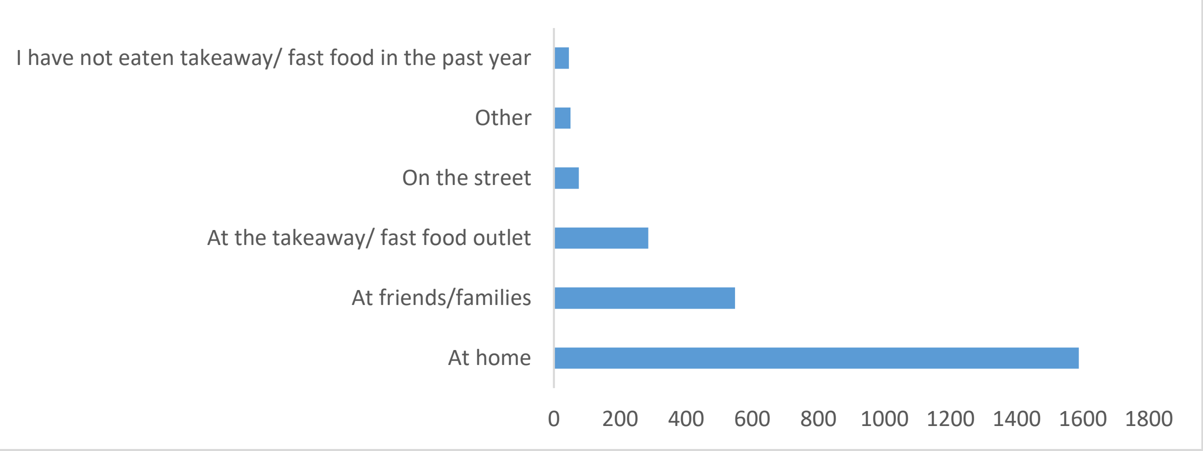


Figure 5-10 Where do you eat takeaway/fast food? (Please tick all that apply) (n = 1724)

Most participants consumed takeaway food with their family (n = 1434) or friends (n = 755) and 330 participants reported consuming takeaway food alone (Fig. 5.11). Other company whilst consuming takeaway food included partner (n = 45), work/colleagues (8), homeless people (1), and the dog (1).

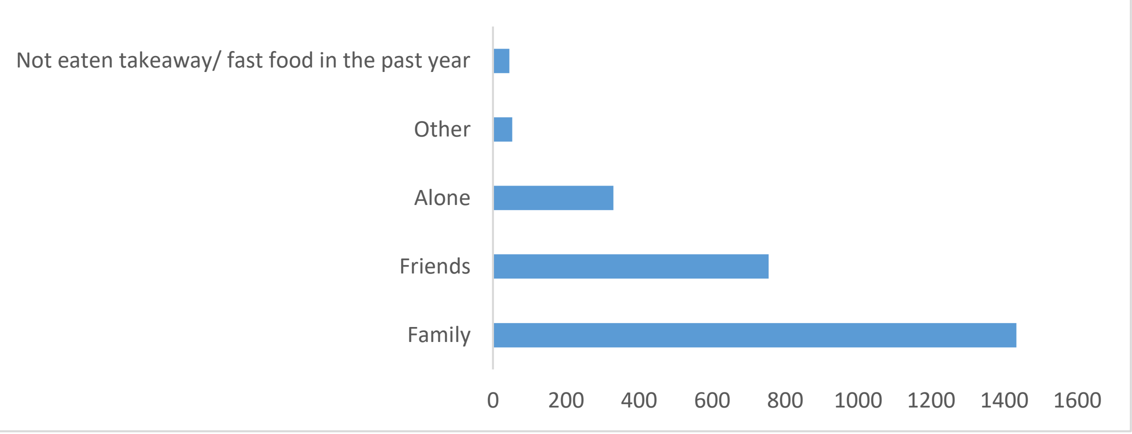


Figure 5-11 Who do you eat your takeaway/fast food with? (Please tick all that apply) (n = 1724)

5.4.5 DIETARY PATTERNS OF A MERSEYSIDE POPULATION

A PCA on 38 food groups was used to analyse habitual and takeaway food intake of 1724 participants. The suitability of PCA was assessed prior to analysis. Inspection of the correlation matrix showed that most variables had at least one correlation coefficient > 0.25, those that did not have a correlation of > 0.25 were removed from the analysis (red meat, fish, nuts and alcohol), leaving a remainder of 34 food groups to re-test. The overall Kaiser-Meyer-Olkin (KMO) measure was 0.83 with individual KMO measures all greater than 0.7, classifications of 'meritorious' to ‘marvelous’ according to Kaiser (Kaiser, 1974). Bartlett's Test of Sphericity (Bartlett, 1954) reached statistical significance ($p < 0.0005$), supporting the factorability of the correlation matrix.

PCA revealed ten components that had eigenvalues > 1, which explained 15.9%, 8.2%, 5.6%, 5.0%, 4.0%, 3.8%, 3.4%, 3.2%, 3.1% and 3.0% of the total variance, respectively. Visual inspection of the scree plot (Appendix 9.25) indicated that four components should be retained (Cattell, 1966). In addition, a four-component solution met the interpretability criterion. As such, four components were retained and a forced factor extraction was run. The four-component solution explained 34.7% of the total variance.

To aid the interpretation of these four components an orthogonal (varimax) rotation was performed. The rotated solution revealed the presence of a number of high positive factor loadings (Table 5.10). The first component in the rotated structure matrix could be described as a ‘Western’ pattern with high positive loadings (> 0.3) obtained for biscuits (0.755), chocolate/confectionary (0.683), buns/cakes/pastries (0.676), fried potatoes (0.585), sugar confectionary (0.581), dairy desserts (0.509), refined grains (0.486), sugar/preserves (0.441), pizza (at home) (0.368), butter/spreads/oil (0.367), SSB (0.328), other potatoes (0.321) and soups and savoury sauces (0.301). There were no negative loadings > −0.3 (Table 5.10).

In the second component it was evident that the high positive loadings (> 0.3) included vegetables (0.754), salad (0.692), fruit (0.624), wholegrains/meal (0.479), yoghurt (0.470), eggs (0.418), legumes (0.415), soups and savoury sauces (0.357), other potatoes (0.316), tea coffee water diet drinks (0.308), cheese (0.300) and no negative loadings >−0.3 (Table 5.10). Thus, this pattern was described as 'Prudent'.

The third component was characterised by high positive loadings (> 0.3) for all of the takeaway food; Chinese (0.663), English (0.639), fast food (0.603), kebab (0.564), pizza (0.549) and Indian (0.482), along with pizza at home (0.329) and sausages (0.317) (Table 5.10). There were no negative loadings > −0.3. This pattern was described as the 'Takeaway Food' pattern.

Finally, the fourth component identified as a 'Meat and Dairy' pattern presented high positive loadings (> 0.3) for milk (dairy) (0.596), bacon/ham (0.515), sausages (0.472), tea/coffee/water/diet drinks (0.438), sugar/preserves (0.398), butter/spreads/oil (0.367) and processed meat (0.364) (Table 5.10). There was a negative loading for other milk (non-dairy) (−0.481). Component loadings of the component matrix are presented in the below table.

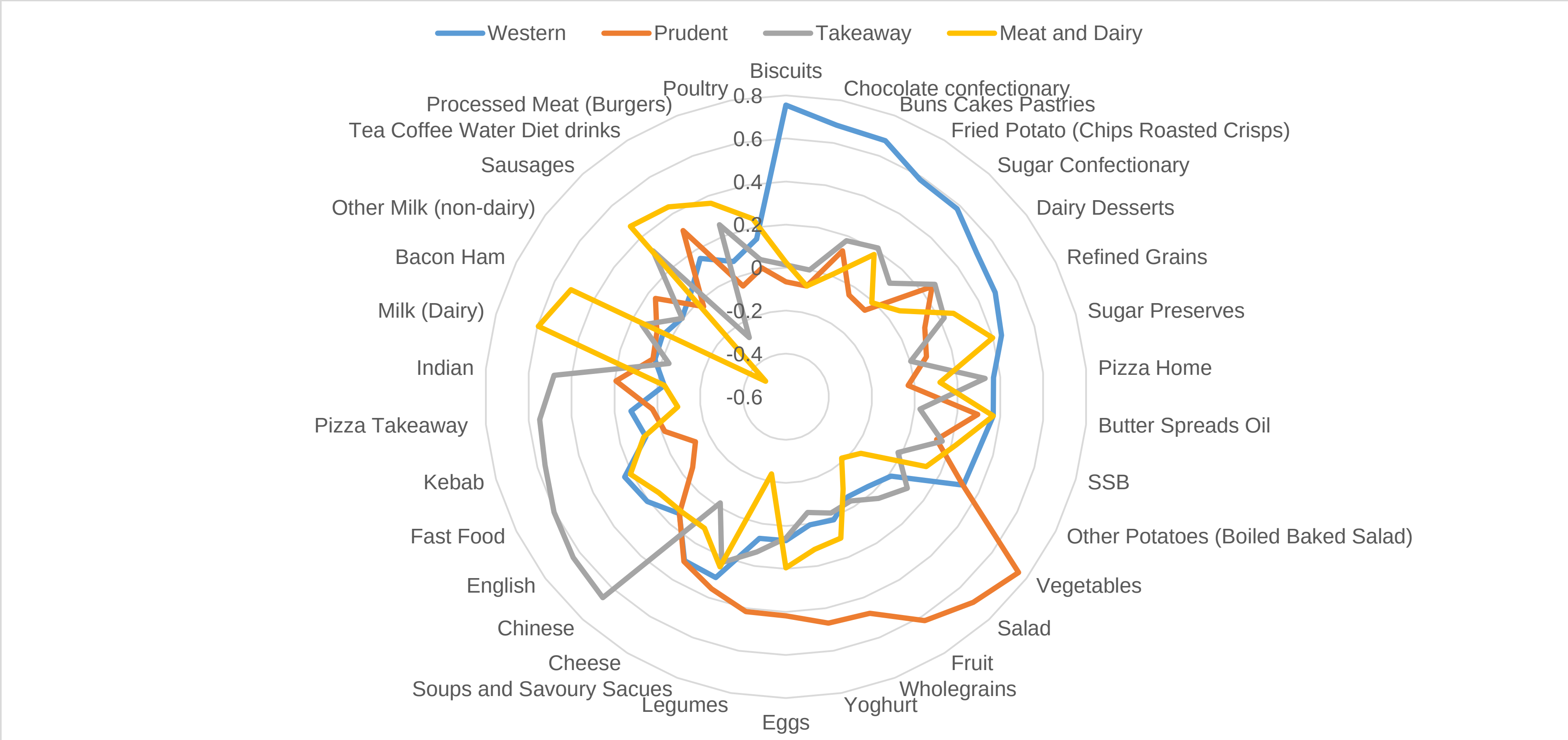
Table 5-10 Rotated component loadings derived from PCA with Varimax rotation for dietary variables that constitute the four main dietary patterns identified

Food Group	Principal Component (Dietary Pattern)			
	C 1	C 2	C 3	C 4
Biscuits	0.755	−0.066	0.012	0.024
Chocolate confectionary	0.683	−0.077	−0.001	−0.077
Buns/Cakes/Pastries	0.676	0.127	0.177	0.012
Fried potato (Chips/Roasted/Crisps)	0.585	−0.044	0.212	0.178
Sugar confectionary	0.581	−0.057	0.114	−0.009
Dairy desserts	0.509	0.250	0.267	0.062
Refined grains	0.486	0.120	0.221	0.269
Sugar preserves	0.441	0.079	0.002	0.398
Pizza (at home)	0.368	−0.030	0.329	0.119
Butter/Spreads/Oil	0.367	0.295	0.025	0.367
SSB	0.328	0.128	0.155	0.220
Other potatoes (Boiled/Baked/Salad)	0.321	0.316	−0.019	0.127
Vegetables	0.012	0.754	0.106	−0.164
Salad	−0.036	0.692	0.039	−0.215
Fruit	−0.054	0.624	−0.030	−0.095
Wholegrains/Wholemeal	0.013	0.479	−0.021	0.104
Yoghurt	0.005	0.470	−0.053	0.121
Eggs	0.068	0.418	0.057	0.195
Legumes	0.069	0.415	0.134	−0.235
Soups and savoury sauces	0.301	0.357	0.227	0.247
Cheese	0.295	0.300	−0.020	0.119
Chinese	0.135	0.134	0.663	0.122
English	0.208	−0.056	0.639	0.140
Fast food	0.237	−0.130	0.603	0.207
Kebab	0.077	−0.014	0.564	0.087
Pizza takeaway	0.123	0.023	0.549	−0.095

Indian	−0.032	0.193	0.482	−0.033
Milk (dairy)	0.033	0.042	−0.035	0.596
Bacon/Ham	0.041	0.070	0.148	0.515
Other milk (non-dairy)	0.005	0.159	0.004	−0.481
Sausages	0.056	−0.033	0.317	0.472
Tea/Coffee/Water/Diet drinks	0.156	0.308	−0.277	0.438
Processed meat (burgers)	0.074	−0.048	0.257	0.364
Poultry	0.145	0.008	0.049	0.242

NB: Major loadings for each item are bolded (factor loadings > 0.3 or < −0.3 are considered significant). Extraction method: Principal component analysis. Rotation method: Varimax.

The component scores for each dietary pattern were inputted into a radar graph to provide a visual interpretation of the differences between the four dietary patterns produced through PCA (Fig. 5.12).



NB: Blue line = the 'Western' dietary pattern (DP1); orange line = the 'Prudent' pattern (DP2); grey line = the 'Takeaway Food' pattern (DP3); yellow line = the 'Meat and Dairy' pattern (DP4).

Figure 5-12 Radar graph of dietary patterns and factor loadings derived through PCA on 34 FFQ food groups (n = 1724)

5.4.6 Socio-demographic and lifestyle determinants

Spearman's correlation was run to assess the relationship between the Western (DP1), Prudent (DP2), Takeaway Food (DP3) and Meat and Dairy (DP4) dietary patterns with the socio-demographic and lifestyle factors (Table 5.11). There was a significant ($p < 0.005$) small positive correlation between the Western pattern and children in the household ($r = 0.12$) and poorer health status ($r = 0.15$), and a significant ($p < 0.05$) small negative correlation with older age ($r = –0.12$), higher educational status ($r = –0.07$), alcohol consumption ($r = –0.11$), and moderate and vigorous exercise ($r = –0.05$). There was a significant ($p < 0.05$) small positive correlation between the Prudent pattern and older age ($r = 0.20$), higher educational status ($r = 0.15$), alcohol consumption ($r = 0.06$) and moderate and vigorous exercise (0.11), and a significant ($p < 0.005$) small negative correlation with poorer health status ($r = –0.15$), increased smoking ($r = –0.10$) and increased daily work activity ($r = –0.06$) (Table 5.11). The Takeaway Food pattern had a significant ($p < 0.05$) small positive correlation with adults in the household ($r = 0.08$), poorer health status ($r = 0.19$), increased smoking ($r = 0.06$), alcohol consumption ($r = 0.06$), increased daily work activity ($r = 0.07$) and moderate and vigorous exercise ($r = 0.11$). Additionally, there was a significant ($p < 0.005$) moderate negative correlation with older age group ($r = –0.33$) and small negative correlation with higher educational status ($r = –0.17$) (Table 5.11). The final Meat and Dairy pattern had a significant ($p < 0.005$) small positive correlation with older age group ($r = 0.08$), children in the household ($r = 0.07$), poorer health status ($r = 0.09$), increased smoking (0.20), increased BMI ($r = 0.12$) and increased daily work activity ($r = 0.07$), and a significant ($p < 0.005$) small negative correlation with higher educational status ($r = –0.17$) (Table 5.11).

Table 5-11 Spearman's correlation for dietary patterns and socio-demographic and lifestyle variables

Variables	DP1	DP2	DP3	DP4	5	6	7	8	9	10	11	12	13	14	15	16
1. DP1	1															
2. DP2	-.14**	1														
3. DP3	.00	-.13**	1													
4. DP4	.02	0.3	-.08**	1												
5. Age group	-.12**	.20**	-.33**	.08**	1											
6. Adults in household	.00	-.00	.08**	.01	-.17**	1										
7. Children in household	.12**	.00	.03	.07**	.03	-.10**	1									
8. Education	-.07**	.15**	.-13**	-.17**	-.02	-.04	-.11**	1								
9. Health status (very good – very poor)	.15**	-15**	.19**	.09**	-11**	.00	.05*	-.12**	1							
10. Smoking frequency	.03	-.10**	.06*	.20**	-.06*	.08**	.00	-.15**	.09*	1						

11. Alcohol (drinks/week)	-.11**	.06*	.06*	.03	.05*	.05	-.13**	.07*	-.08	.06	1					
12. BMI	.04	.00	.03	.12**	.16**	-.04	.08**	-.13	.34**	-.05*	-.11*	1				
13. WHtR	.01	-.03	.01	-.03	-.01	-.02	-.02	.01	.02	-.00	-.02	-.01	1			
14. Daily work activity	.04	-.06**	.07**	.07**	-.11	.00	.10**	-.12**	-.09**	.11**	-.00	-.08**	-.05	1		
15. LPA (min/day)	.00	.04	.04	.02	-.14**	.02	.07**	-.04	-.08	.04	.03	-.05*	.00	.28**	1	
16. MVPA (min/day)	-.05*	.11**	.11**	.03	-.14**	.02	.04	-.04	-.19**	.03	.02	-.18**	. -.01	26**	.41**	1

NB: **Correlation is significant at the 0.01 level (2-tailed). *Correlation is significant at the 0.05 level (2-tailed). LPA = Low physical activity. MVPA = Moderate and vigorous physical activity.

5.4.7 DIFFERENCES BETWEEN GROUPS

A Mann-Whitney U test determined if there were differences in dietary pattern scores for gender, ethnicity, supplement use and long-term illness (Table 5.12). Males scored significantly ($p < 0.0005$) higher than females on the Meat and Dairy dietary pattern and Takeaway Food dietary pattern, meaning they were more likely to consume those patterns than females (Table 5.12). Males scored significantly ($p < 0.05$) lower than females on the Prudent pattern therefore, were less likely to consume that pattern. Participants from non-white ethnicities scored significantly ($p > 0.05$) higher than white ethnicities on the Takeaway dietary pattern and Prudent dietary pattern, meaning they were more likely to consume those patterns (Table 5.12). Whereas non-whites scored significantly ($p < 0.0005$) lower than whites on the Meat and Dairy pattern and were therefore less likely to consume that pattern. Non-supplement users scored significantly ($p < 0.0005$) lower than supplement users on the Prudent pattern meaning they were less likely to consume that pattern and supplement users scored significantly ($p < 0.005$) lower than non-supplement users on the Takeaway and Meat and Dairy patterns; suggesting that they were less likely to consume both patterns (Table 5.12). For long-term illness, those responding yes scored significantly ($p < 0.05$) lower than those responding no on the Takeaway pattern and were therefore less likely to consume that pattern than those with no illness (Table 5.12).

Table 5-12 Mann-Whitney U tests for differences between dietary patterns and socio-demographic or lifestyle variables

Variables	Dietary patterns from PCA			
	DP1 (Western)	DP2 (Prudent)	DP3 (Takeaway)	DP4 (Meat and Dairy)
Gender (n = 1721)				
Male	−0.300 (−0.593–0.276)	−0.307 (−0.720–0.345)	−0.022 (−0.463–0.554)	0.222 (−0.322–0.804)
Female	−0.219 (−0.554–0.243)	−0.142 (−0.633–0.486)	−0.193 (−0.550–0.218)	−0.079 (−0.541–0.435)
p-value	0.292	0.013	< 0.0005	< 0.0005
Ethnicity (n = 1724)				
White	−0.226 (−0.556–0.246)	−0.170 (−0.653–0.439)	−0.183 (−0.547–0.238)	−0.005 (−0.491–0.529)
Non-White	−0.334 (−0.557–1.187)	0.079 (−0.557–1.187)	0.042 (−0.450–0.741)	−0.451 (−0.888–0.039)
p-value	0.190	0.017	0.001	< 0.0005
Supplements (n = 1689)				
Yes	−0.258 (−0.563–0.199)	−0.002 (−0.504–0.650)	−0.224 (−0.576–0.158)	−0.119 (−0.670–0.442)
No	−0.215 (−0.552–0.299)	−0.289 (−0.742–0.332)	−0.142 (−0.515–0.342)	0.035 (−0.407–0.035)
p-value	0.108	< 0.0005	0.002	< 0.0005
Illness (n = 1724)				
Yes	−0.168 (−0.545–0.320)	−0.154 (−0.688–0.546)	−0.226 (−0.583–0.162)	0.060 (−0.512–0.631)
No	−0.261 (−0.560–0.202)	−0.170 (−0.630–0.439)	−0.155 (−0.518–0.302)	−0.061 (−0.502–0.475)
p-value	0.138	0.846	0.042	0.117

NB: All values are median factor loadings unless otherwise stated. Interquartile range (25%–75%).

A Kruskal-Wallis H test showed the difference between groups of variables with more than two categories for the four dietary patterns (Table 5.13). For marital status, the divorced/widowed or separated group scored significantly ($p < 0.05$) lower on the Western dietary pattern and the Takeaway pattern and were therefore less likely to consume these patterns, and scored significantly ($p < 0.005$) higher on the Meat and Dairy pattern (Table 5.13). However, those in the single group scored significantly ($p < 0.0005$) lower and were less likely to consume the Prudent dietary pattern than those who were married or divorced/separated/widowed. Retired participants scored significantly ($p < 0.005$) lower and were less likely to consume the Western and Takeaway dietary patterns than the employed, unemployed, students or other. Whereas the other group (ill health, disability, carer or maternity leave) scored significantly ($p < 0.0005$) higher and were more likely to consume the Prudent and Meat and Dairy dietary patterns than the other groups (Table 5.13).

Table 5-13 Kruskal-Wallis H tests for dietary patterns and background variables

Variables	Dietary patterns from PCA			
	DP1 (Western)	DP2 (Prudent)	DP3 (Takeaway)	DP4 (Meat and Dairy)
Marital Status				
Single	−0.194 (−0.538–0.320)	−0.296 (−0.763–0.273)	−0.059 (−0.442–0.405)	−0.079 (−0.556–0.461)
Married	−0.272 (−0.581–0.192)	−0.029 (−0.514–0.580)	−0.252 (−0.578–0.129)	0.019 (−0.476–0.529)
Divorced	−0.328 (−0.596–0.173)	−0.128 (−0.651–0.589)	−0.453 (−0.724–−0.010)	0.143 (−0.382–0.635)
p-value	0.012	< 0.0005	< 0.0005	0.006
Employment Status				
Employed	−0.231 (−0.558–0.222)	−0.154 (−0.612–0.412)	−0.188 (−0.534–0.266)	−0.048 (−0.490–0.459)
Unemployed	−0.146 (−0.544–0.367)	−0.370 (−0.947–0.404)	−0.078 (−0.479–0.437)	0.202 (−0.458–0.821)
Student	−0.268 (−0.540–0.292)	−0.269 (−0.703–0.437)	−0.078 (−0.458–0.306)	−0.182 (−0.618–0.293)
Retired	−0.486 (−0.696–−0.013)	0.101 (−0.393–0.0.679)	−0.572 (−0.884–−0.183)	0.109 (−0.399–0.667)
Other*	−0.117 (−0.423–0.096)	0.174 (−0.755–1.006)	−0.035 (−383–0.492)	0.235 (−0.684–0.891)
p-value	0.005	< 0.0005	< 0.0005	< 0.0005
NS-SEC				
Managerial	−0.284 (−0.592–0.176)	−0.048 (−0.476–0.499)	−0.262 (−0.616–0.082)	−0.092 (−0.512–0.340)
Intermediate	−0.289 (−0.577–0.213)	−0.148 (−650–0.383)	−0.187 (−0.543–0.286)	−0.043 (−0.495–0.440)
Manual	−0.173 (−0.504–0.302)	−0.370 (−0.807–0.409)	−0.049 (−0.418–0.437)	0.090 (−0.399–0.678)
Unemployed	−0.213 (−0.549–0.299)	−0.219 (−0.742–0.460)	−0.097 (−0.516–0.352)	−0.048 (−0.596–0.535)
p-value	0.057	< 0.0005	< 0.0005	0.001

NB: All values are median factor loadings unless otherwise stated. Interquartile range (25%–75%). *Other = ill health, disability, carer or maternity leave. NS-SEC: National Statistics Socio-economic Classifications.

5.5 DISCUSSION

5.5.1 TAKEAWAY FOOD CONSUMPTION

The present study aimed to investigate takeaway food consumption within wider dietary patterns of the study population and their self-reported socio-demographic and lifestyle determinants. The findings showed that takeaway food consumption was higher in the participant sample than in the pilot study sample, with 37% of participants consuming takeaway food once per week or more compared to 23% of participants previously (Fig. 5.2). This was expected due to the inclusion criteria (consumption of takeaway or fast food at least once in the last year). Consumption rates were also higher than the FSA report (Food Standards Agency, 2014) showing that 27% of respondents in England consumed takeaway food at least once in the previous seven days and the Public Health England estimate of 27.1% of the population (Public Health England, 2017a) (Chapter 4). However, in Australia, it was reported that over one-third (37.7%) of adults reported consumption of takeaway foods once per week or more and 33.6% consumed takeaway food 1–3 times per month (Miura et al., 2012). Moreover, consumption of fast food in American adults exceeded all findings, with a reported mean consumption of 3.9 fast food meals per week from the US National Health and Nutrition Examination Survey (NHANES) 2005–2010 data (Kant et al., 2015).

Takeaway food intake from aggregated FFQ data (Fig. 5.3) produced higher consumption results than self-reported totals. From the totalled FFQ food group responses, 9% consumed takeaways less than 3 times per month and 91% consumed takeaway food at least once per week, with 59% consuming at least one portion per day. These higher estimates of consumption suggest that the

FFQ method may have overestimated consumption and illustrates the rationale for needing a more accurate measurement of dietary intake; and why a biomarker for self-reported takeaway food may be advantageous (Study 3).

Again, Chinese ranked as the most widely consumed takeaway food in the study sample (Fig. 5.4), supporting the pilot findings (Fig. 4.2, Chapter 4). Fast food closely followed as second for consumption, replacing pizza in the pilot study and Indian in 3rd position, replacing fish & chips in the pilot. Takeaway food purchasing data from the UK for 2006, 2008, 2010, 2012, 2014 and 2016 has shown that pizza, Chinese and Indian have consistently remained in the top 3 favourite positions (Just Eat, 2017). Nevertheless, as mentioned previously, data from a food sector report may lack academic rigour. Therefore, review with caution.

5.5.1.1 Socio-demographic and lifestyle determinants of takeaway food

When looking at the median consumption of takeaway food and their socio-demographic determinants, men consumed significantly ($p = 0.001$) more takeaway food than women (Table 5.7), comparable to previous research in Australia (Smith et al., 2009; Trapp et al., 2015), Europe (Orfanos et al., 2009), New Zealand (Smith et al., 2014), and the US (Kant et al., 2015; Wade, 2018). However, these results were different to those found in a UK study where the only gender difference was that boys consumed takeaway food at home more than girls (Adams et al., 2015). An explanation for this difference between the two UK studies was that the study design and focus were dissimilar. The participant sample in the current project were self-reported takeaway food consumers (consumption > 1/year) whereas the study by Adams et al. (2015) used secondary food consumption data from the NDNS on the general population. Moreover, the study location of Merseyside has higher rates of takeaway food consumption than other areas of the UK (UKCRC Centre for Diet and Activity Research (CEDAR), 2017b) inferring that the study population may have different takeaway food behaviours to populations in other cities and towns in the UK. Concordant with the results from the present study, a recent UK report found that men were more likely than women (59% compared with 50%) to have consumed takeaway food from a takeaway outlet or restaurant in the previous month (Food Standards Agency, 2017). However, the report did not examine the types of takeaway food consumed and whether there was a gender variation according to type. In the current study, further detail was obtained by segregating each takeaway food category and found that males consumed significantly more fast food ($p = 0.003$), English ($p = 0.014$), pizza ($p = 0.024$) and kebab ($p < 0.0005$) takeaways than females (Table 5.8). There were no significant differences between genders for Indian ($p = 0.849$) or Chinese ($p = 0.120$) takeaways. Previous studies in the UK have focussed on takeaway food as a whole and this level of detail has been absent to make comparisons (Adams et al., 2015). In the New Zealand Adult Nutrition Survey comparisons were made between sexes for 12 fast foods groups and found that females were significantly ($p < 0.05$) more likely to consume vegetables (including salads/coleslaw) than males (Smith et al., 2014); indicative of healthier choices compared with males. This finding, while preliminary, suggests that the higher consumption of fast food, English, pizza and kebab in males is due to increased demand for on the go convenience food.

Whereas Chinese and Indian meals tend to be consumed during social occasions among family and friends (Fig. 5.11).

Consumption of total takeaway was significantly ($p < 0.0005$) higher among the youngest age groups (18–25 and 26–35 years) and decreased steadily as age increased (Fig. 5.5), in agreement with the previous UK study on NDNS data (Adams et al., 2015). In other nations, the peak consumption age for takeaways is from 18 to 29 years (van der Horst et al., 2011; Smith et al., 2014; Kant et al., 2015). This was of particular relevance for the fast food ($r = -0.34$, $p < 0.01$) and pizza ($r = -0.40$, $p < 0.01$) categories (Table 5.6). Fast food has been suggested to be a symbol of youth identity and part of youth culture (Bugge, 2011) with increased consumption linked to limited cooking skills (Hartmann et al., 2013) and peer influence (Blow et al., 2017). Moreover, the price of food outside of the home was considered the most important factor for young respondents (aged 16–24) in the UK when deciding on food outlet (18%, compared with 4%–12% in older groups) (Food Standards Agency, 2017). All age groups consumed Indian takeaway food and there were no correlations with age.

Ethnicity was a predictor for some takeaway food categories with non-white participants consuming significantly ($p < 0.05$) more fast food and pizza than white participants, but not for total takeaway food ($p = 0.105$) (Table 5.8) . A recent study on fast food intake in 18–25 years old white and African Americans found that the feminine gender orientation and education serve as protective factors, while African American race and male sex serve as risk factors (Wade, 2018), comparable to the findings in the present study. Non-whites in Texas exhibited higher obesity rates and increased accessibility and consumption of fast food (Dunn et al., 2012).

In relation to socio-economic status, education was a strong predictor of takeaway food consumption with the least educated participants consuming significantly ($p < 0.01$) more fast food, kebab, pizza, Chinese, and English takeaways than those with a higher education (Table 5.6). Similar results were noted by Thornton et al. (2010) and Burgoine et al. (2016) for fast food consumption and lower educational levels. Conversely, the reverse was shown for education and Indian takeaway food, with higher educated participants consuming more Indian takeaway than those with a lower education ($r = 0.02$), however these results were non-significant (Table 5.6). An Australian study that aggregated 22 takeaway foods into ‘‘less healthy’’ and ‘‘healthy’’ choices, based on the Australian Guide to Healthy Eating, investigated the socio-economic differences in takeaway food and found that the least educated regularly consumed more potato chips, savoury pies, fried chicken and non-diet soft drinks and were less likely to consume curry (Miura et al., 2012). Education appears to be a protective factor for takeaway food consumption; however, this may be attributed to having a higher socio-economic status and the associated characteristics such as increased economic resources (Burgoine et al., 2016).

In the current study, employment status and NS-SEC (occupational class) were strong determinants of takeaway food consumption (Table 5.9). Retired respondents consumed significantly ($p < 0.015$) less takeaway food on all categories, concordant with the FSA report showing that the retired group were less likely than other groups to have consumed takeaways (23% compared with 57%–64% of those with other economic statuses) (Food Standards Agency, 2017). Reasons for this could be that some retired individuals have not developed a habit of eating takeaway foods during earlier years of life and may have less disposable income (Adams et al., 2015). Participants in the manual occupational class consumed significantly more fast food, Chinese, English and pizza than the other occupational classes (Table 5.9). An explanation for this may be that those in the lowest socio-economic groups display less food and nutrition knowledge (Parmenter et al., 2000), less confidence with cooking (Adams and White, 2015), fewer food preparation facilities (Appelhans et al., 2014) and lower disposable income (Adam, 2009). Moreover, previous research has shown that deprived areas of the UK also exhibit higher densities of takeaway food outlets (Maguire et al., 2015). A former study found the only difference for occupational class in the UK was that adults in higher NS-SEC were more likely to eat meals out at least once per week (Adams et al., 2015); suggesting that they are better able to afford foods from restaurants and cafe´s but do not differentiate what meals were eaten out.

The biggest predictor of takeaway food consumption in terms of marital status was for those who were single, specifically for fast food and pizza compared to married or divorced participants (Table 5.9). A possible explanation for these results may be that those who are single are often a younger age and it has already been reported that the younger age groups consume more takeaway overall. Living with children was a determinant ($p < 0.005$) of takeaway food consumption in the current study (Table 5.5). The present finding seems to be consistent with other research which found that respondents with children aged under 16 in the household were more likely to have consumed takeaway food (68% compared with 49% of those in adult-only households) (Food Standards Agency, 2017).

As expected, takeaway food intake was associated with increased self-reported BMI (Table 5.5) given to their typically energy-dense nutritional content (Jaworowska et al., 2014). Use of fast food outlets in adults in the UK NDNS was significantly associated with higher odds of obesity (Penney et al., 2017). These findings corroborate previous research showing that frequent takeaway food consumption (> 2 times per week) predicted 5 year weight gain In Australian adults (Smith et al., 2017). That said, a recent study on the behavioural patterns of Canadian adolescents and concurrent BMI status suggested that the greatest predictor of obesity issues in adulthood could be a combination of unhealthy behaviours including alcohol, smoking and sedentary lifestyle (Laxer et al., 2018). In the current study, there were no correlations between waist-to-height ratio and takeaway food category (Table 5.5). A speculated reason for this is that many of the participants with a higher BMI reported that they did not know their waist size or did not want to provide this information; only 1000 (724 missing) provided waist size information.

Findings from the present study indicated that smoking was positively ($p < 0.01$) associated with increased consumption of fast food, Chinese, English, pizza and kebab (Table 5.6). Alcohol was positively ($p < 0.01$) associated with consumption of Chinese and Indian takeaway food (Table 5.6). Townsend (2017) stated that in the UK drinking at home has increased in recent decades as a consequence of increased availability and relatively low cost. Moreover, links were made between potentially unhealthy behaviours including drinking alcohol and fast food consumption (Townshend, 2017). Therefore, it seems possible that the correlation between Chinese or Indian takeaway food and alcohol consumption in the current study are due to consumption in the home (Fig. 5.10).

Participants reporting a poorer health status also consumed increased takeaway food consumption, specifically fast food, Chinese, English, pizza and kebab (Table 5.6).

Increased sedentary lifestyles including prolonged screen time have been associated with increased takeaway food consumption (Smith et al., 2009). However, in the present study increased daily occupational activity had a positive correlation with total takeaway food consumption, specifically fast food, Chinese, English and pizza but no correlation with Indian or kebab (Table 5.6; Fig. 5.8). Moreover, there was a significant but small positive correlation between low physical activity and fast food and no correlation with Chinese, Indian, English, pizza or kebab (Table 5.6). Suggesting that low physical activity may be associated with being a younger age male. Moderate and vigorous physical activity was significantly positively correlated with fast food, Chinese, pizza and kebab, with no correlations for Indian or English takeaways (Table 5.6). Indian takeaway had the least correlations with other socio-demographic and lifestyle variables, suggesting consumption of this takeaway food to be widespread among the population and less homogeneity of the consumers.

5.5.1.2 TAKEAWAY FOOD CONSUMER BEHAVIOURS/PSYCHO-SOCIAL

Several themes emerged regarding the self-reported reasons for takeaway food consumption. Satisfaction was the strongest theme to arise as participants described takeaway food as ‘a treat’, that they ‘liked the taste’, to eat ‘something different’ and ‘they are cheap’ (Fig. 5.9). Notions of convenience were also described through answers including takeaway food being ‘easily available’ and ‘available close to my home’, ‘too busy to cook or ‘can’t be bothered cooking’ (Fig. 5.9). The third was a social theme from the responses ‘for social occasions’ and ‘my friends/family like it’. Themes of lower significance were regarding preparation and facilities including reports that participants ‘don't like to prepare food’, ‘don’t know how to cook’, ‘no facilities’ and like having ‘no dishes to clean’ (Fig. 5.9). Finally, other reasons covered the physical and emotional theme related to consumption of takeaways; ‘being hungover’, ‘health issues that limit cooking’, ‘disability’, ‘tired’, ‘stressed’, ‘comfort’ and ‘bored’ (Fig. 5.9). Taste has been a fundamental determinant of highly palatable foods such as fast food (Glanz et al., 1998). A recent qualitative study exploring ‘chicken’ shops in a low socio-economic area in London found that participants particularly liked the community status of such outlets and described the food as tempting, addictive, delicious and affordable (Thompson et al., 2018).

Previous research on food purchasing behaviours in New Zealand reported that out-of-home meals were generally purchased by participants that wanted a break from cooking, lacked the time, energy, or the ingredients to prepare a meal, hoped to provide a treat for their family, or wanted to celebrate an occasion (Maubach et al., 2009). In the current study, most participants consumed their takeaways at home or at friends/families (Fig. 5.10) and with family/friends (Fig. 5.11). The findings from a study on Australian adults suggested that fast food consumption was influenced by the convenience, the satisfaction, family and friends, and facilitating factors (such as busy lifestyles, experiencing cravings for fast food, not knowing how to cook, working long hours, and eating alone) (Dunn et al., 2011). Convenience, social interaction and a general demand for tasty meals are reoccurring factors when researching the perceived reasons for takeaway food consumption and limited attention is given to the long-term health implications.

5.5.2 DIETARY PATTERNS OF TAKEAWAY FOOD CONSUMERS AND ASSOCIATED SOCIO-DEMOGRAPHIC AND LIFESTYLE DETERMINANTS

In this study, four patterns of diet were identified using PCA in adults aged 18–64 years (n = 1724). The 'Western' dietary pattern was the most prevalent in the population, accounting for 15.9% of the total variance in the dietary intake, followed by a 'Prudent' pattern accounting for 8.2 %, the 'Takeaway Food' pattern accounting for 5.6%, and finally a 'Meat and Dairy' pattern accounting for 5.0% (Table 5.10). Variables such as age, gender, ethnicity and anthropometrics and dietary pattern scores determined relationships or difference between groups.

5.5.2.1 WESTERN PATTERN

The first and most prevalent pattern had high component scores (> 0.3) for many western foods (including biscuits, chocolate confectionary, buns/cakes/pastries, fried potatoes, sugar confectionary, dairy desserts, refined grains, sugar/preserves, pizza (at home), butter/spreads/oil, SSB, other potatoes and soups and savoury sauces (Fig. 5.12). The majority of these foods are characterised as having a high-fat, high-sugar, and excess salt content (Manzel et al., 2014). A similar pattern was recently determined from the UK NDNS data and was labelled 'Snacks, fast food, fizzy drinks' accounting for 3.9% of the variance in the population's dietary intake (Roberts et al., 2018). Many of the foods and drinks consumed within the Western type dietary pattern could be considered as ultra-processed; industrial formulations manufactured mostly or entirely from ingredients derived from constituents of foods and additives. Examples of ultra-processed foods include packaged breads, buns, pizzas and cakes, crisps, industrially-produced desserts, fizzy drinks, fish and chicken nuggets, instant noodles and soups, and frozen ready meals (Fiolet et al., 2018). Results from a study of 19 European countries showed that 50.7% of the UK diet came from ultra-processed foods, more than any other European country (Monteiro et al., 2018). Extensive research on 74, 470 participants in the NutriNet-Santé cohort showed that those consuming the highest amounts of ultra-processed foods also consumed higher amounts of sweet products and soft drinks (Julia et al., 2018) which is concordant with the findings in this study. The consequences of consuming more ultra-processed foods, evidenced by a large prospective study of 104,980 healthy French adults, include a significant increase in risk of overall cancer and breast cancer (Fiolet et al., 2018). Moreover, consumption of an unhealthy/western dietary pattern may increase CVD and mortality risk (Hamer et al., 2010; Atkins et al., 2016).

In the current study, the Western type dietary pattern was positively ($p < 0.005$) associated with younger age, more children in the household, poorer health status, lower education, and single marital status (Table 5.11; 5.13). Similarly, in the recent French study, higher consumption of ultra-processed foods were independently associated with male gender, younger age, lower education, smoking, and overweight and obesity (Julia et al., 2018). In the Balearic islands' adolescents, del Mar Biboloni (2012) showed that a Western dietary pattern was associated with low parental socio-economic status, considerable leisure-time on sedentary behaviours (such as media screen time) and body image. Consumption of a Western dietary pattern was significantly associated with a higher prevalence of overweight/obesity in US children, independent of fast food intake (Poti et al., 2014). Moreover, a recent review concluded that a Western dietary pattern accentuated the association of polymorphisms with the metabolic syndrome (Hosseini-Esfahani et al., 2018).

The present study found that being retired, increased alcohol consumption and moderate/vigorous exercise served as protective factors against the consumption of the Western dietary pattern (Table 5.11; 5.13). This could be a result of older respondents not consuming as much western type foods or more 'health conscious' individuals choosing to consuming healthier dietary patterns.

5.5.2.2 Prudent pattern

A healthier pattern was the second dietary pattern to emerge from the PCA and included high component scores (> 0.3) for other potatoes (boiled, baked, salad), vegetables, salad, fruit, wholegrains, yoghurt, eggs, legumes, soups and savoury sauces, and cheese) (Fig. 5.12). In the context of the dietary pattern literature this pattern was most similar to a 'Prudent dietary pattern', as identified in the Caerphilly Prospective Cohort Study (Mertens et al., 2017) and the Nurses' Health Study (Heidemann et al., 2008) and several other studies (Bédard et al., 2015; Atkins et al., 2016). The Prudent pattern was also comparable to the Mediterranean dietary pattern; components included vegetables, legumes, fruits, nuts, whole grains, fish, monounsaturated fat–saturated fat ratio, alcohol, and meat (Fig. 5.12). A dietary pattern comprising these foods have been praised for having positive effects on longevity (Mitrou et al., 2007), health outcomes (Kant, 2004) and wellbeing (Lai et al., 2014). Roberts et al. (2018) also identified a similar pattern in UK adults in the NDNS and classified it as the ‘Fruit, vegetables, oily fish’ pattern. A healthy/prudent dietary pattern has continually shown inverse associations with CVD and mortality risk (Newby and Tucker, 2004; Hou et al., 2015; Li et al., 2015; Rodriguez-Monforte et al., 2015; Atkins et al., 2016).

In the current study, adherence to the Prudent dietary pattern was positively ($p < 0.05$) associated with female gender, older age, higher education, increased alcohol consumption, increased moderate/vigorous exercise, consumption of supplements, non-white ethnicities, and sedentary work (Table 5.11; 5.12; 5.13) . Similarly, the ‘fruit, vegetables, oily fish’ pattern from NDNS was positively ($p < 0.01$) associated with being non-white, age, income, and socio-economic classification (NS-SEC) (Roberts et al., 2018). Participants in the present study that were less likely to consume the Prudent dietary pattern were single, unemployed, working in routine/manual occupations, smokers or had poor health status (Table 5.11; 5.12; 5.13). Whereas in the NDNS the ‘fruit, vegetables, oily fish’ pattern was negatively associated with male gender, smoking and BMI. These findings suggest some homogeneity among the wider UK population and thus the participants in this study may be representative of the general population.

5.5.2.3 TAKEAWAY FOOD PATTERN

For the third dietary pattern, all takeaway foods correlated with one another; presenting high factor loadings (> 0.3) for Chinese, English, fast food, kebab, pizza and Indian, along with pizza at home and sausages (Fig. 5.12). In a study on the dietary patterns of US children, those that were low or high fast food consumers were more likely to consume a Western dietary pattern compared with fast food non-consumers (Poti et al., 2014). The remainder of the diet (habitual diet) may be more damaging to health than the takeaway food itself due to the proportion of habitual to out-of-home foods consumed, but when consumed in combination could have an unprecedented detrimental effect on health (Bahadoran et al., 2012; Smith et al., 2012; Donin et al., 2017) .

Consumption of the Takeaway Food dietary pattern was positively ($p < 0.05$) associated with increased adults in the household, poorer health status, smoking, increased alcohol consumption, increased physical daily occupational activity and moderate/vigorous exercise (Table 5.11). A number of the associated behaviours would be considered harmful to health (smoking and alcohol) however; a more physical occupation (manual rather than desk based) and increased time participating in moderate/vigorous exercise may serve as protective factors when consuming takeaway food. Earlier reports of frequent out-of-home food (including fast food) consumption has shown a positive correlation with BMI (Kant et al., 2015) however, results for this pattern were contradictory. Those from an older age and higher education were least likely ($p < 0.005$) to consume the Takeaway Food dietary pattern (Table 5.11); suggesting an age and socio-economic gradient for takeaway food consumption as mentioned previously (Adams et al., 2015; Maguire et al., 2015).

5.5.2.4 MEAT AND DAIRY PATTERN

Individuals scoring highly on the Meat and Dairy pattern presented high factor loadings (> 0.3) for milk (dairy), bacon/ham, sausages, tea/coffee/water/diet drinks, sugar/preserves, butter/spreads/oil and processed meat (Fig. 5.12). There was a negative loading (> −0.3) for other milk (non-dairy). This pattern was laden with saturated fat and processed meats and despite much debate regarding the health risks of saturated fat (Nettleton et al., 2015), it is widely evidenced that processed meats are harmful to health (Etemadi et al., 2017; Crippa et al., 2018; Rosato et al., 2018). A study on older British adults identified a similar ‘high fat/low fibre’ dietary pattern (high in red meat, meat products, white bread, fried potato, eggs) which was associated with an increased risk of all-cause mortality (Atkins et al., 2016). In several other studies, processed meat has been a component in an unhealthy/western dietary pattern (Thorpe et al., 2016; Gibbons et al., 2017).

In the present study, adherence to the Meat and Dairy dietary pattern was positively ($p < 0.005$) associated with older age, more children in the household, poorer health status, smoking, increased BMI, lower education and increased daily occupational activity (Table 5.11). In an Australian study, participants consuming a pattern characterised by higher consumption of red and processed meat and refined grains were also more likely to have a lower level of education, a higher BMI, and smoke however, they did not meet physical activity recommendations (Thorpe et al., 2016). In the UK, physical activity guidelines states that adults 19–64 years should engage in at least 150 minutes (2½ hours) of moderate intensity activity per week (30 minutes on at least 5 days a week) (Department of Health, 2011). It is estimated that 39% of UK adult population, rising to 47% (the highest proportion) in North West England, are physically inactive and failing to meet government recommendations for physical activity (British Heart Foundation, 2017). Therefore, it appears that the current study reporting a MED of 34.3 (IQR 17.1–68.6) minutes per day for moderate/vigorous activity to be higher than anticipated (Table 5.4). It seems possible that these results are due to some over reporting in the current sample, participants may have mislabelled physical activity as moderate/vigorous activity instead of lower physical activity or the sample may not be representative of adults in North West England. This also accords with earlier research on the validity of self-reported physical activity, which showed that self-reported physical activity was on average 84% higher than an objective criterion (Lee et al., 2011); a serious problem for population studies (Rzewnicki et al., 2003).

5.6 STRENGTHS AND LIMITATIONS

The present study had several strengths. First, this was a population-based study with a large sample size of 1724 participants. Second, takeaway food consumption was analysed in detail and included many takeaway food types compared with previous research, which commonly focuses on total takeaway food or 'fast food' (Smith et al., 2012; Poti et al., 2014; Adams et al., 2015; Zagorsky and Smith, 2017). Third, each specific takeaway food category was studied across socio-demographic groups and for individual lifestyle factors.

That said, there were a number of limitations, including that cross-sectional data cannot demonstrate causality. The participant sample may not be entirely representative of self-reporting adults in Merseyside, adults in the UK or further afield. Participants self-select and with no empirical information on non-participants bias cannot be excluded. Individuals over 64 years of age on social media questioned the reason for exclusion and reported that they consumed takeaway food, thus valuable data on older adults may be missing from the present study. Nevertheless, takeaway food consumption did decrease with older age. There was difficulty when estimating the target sample size for the current study as there were a number of aspects to consider including performing a power calculation for prevalence of takeaway food consumers in Merseyside and requiring a large enough sample for dietary pattern analysis, which is somewhat arbitrary, particularly because power calculations are not explained in other studies. Moreover, the study required a large enough sample to recruit a sub-sample of 200 participants for Study 3. A retrospective calculation using the number of participants recruited for Study 3 (n = 154) and the 1724 participant sample in Study 2 (1724/154 x 200) estimated 2,239 participants to reach the 200 target set by AU.

The female to male ratio was uneven and there may have been a female bias in the results; female bias has occurred in similar studies (Barnes et al., 2016); however, there was a large enough male sample (n = 297) in the study to make statistical inferences. Facebook has been said to be an effective method of recruitment of low-income women for nutrition education (Lohse, 2013) suggesting that the method of recruitment may have influenced female participation. In this study, albeit there was a high concentration of females, measures of socio-economic status showed an even distribution.

Self-reported dietary intake and lifestyle information proposes an unknown degree of misreporting. Self-report measures produce measurement error because factors other than the measurement in question will influence how individuals will respond to the measure (Field, 2009). With regards to the FFQ style format of the habitual and takeaway food sections in the EHS, it was difficult to estimate usual intake for an entire year (long-term intake requires memory) and there was a possibility of recall bias where individuals selectively recall food items (as with all self-reported methods). It is suggested that FFQs approximately underreport a third of energy and protein intake (Naska et al., 2017). There are also issues in validating FFQs because an FFQ from one country is unlikely to be appropriate for other populations. However, the EPIC-Norfolk FFQ used for modification in this study had been validated in UK populations (Day et al., 2001; McKeown et al., 2001). Moreover, content and convergent validity were completed for the EHS. In the current study an objective measures of food consumption, i.e. biomarkers were not included.

There was a degree of subjectivity in determining food groups before application of PCA, the removal of food groups based on factor scores < 0.3 or < -0.3, the treatment of the input variables and labelling of dietary patterns. The naming/labelling of dietary patterns is something that is somewhat subjective even when looking at previous research (Pryer et al., 2001; Hosseinzadeh et al., 2016; Cunha et al., 2018). For instance, a traditional dietary pattern among British adults may be very different to a traditional dietary pattern in Iran or Brazil. These patterns would reflect quite different food groups and outcomes. Thus, results would not be representative of other nations and PCA may not derive comparable patterns in different populations. Finally, many of the correlations were small (< 0.3) and thus caution must be applied when discussing associations, a reason for this could be due to the large sample size, heterogeneity among the participants, and the presence of confounding variables.

5.7 Conclusion

There are many factors influencing takeaway food consumption and wider dietary patterns and these vary greatly on an individual basis. This study showed that takeaway food was consumed at varying degrees and was dependant on many socio-demographic and lifestyle determinants. Being male, in a younger age group, having children in the household, alcohol use and smoking were positively associated ($p < 0.05$) with total takeaway food consumption, whereas older age groups and higher qualification were negatively ($p < 0.01$) associated. For specific takeaway food categories, fast food, pizza, Chinese, English and kebab were higher in those with self-reported lower health status, lower education and non-supplement users. Fast food consumption was prevalent in younger age groups, male gender and in lower socio-economic backgrounds whereas Indian takeaway food was consumed homogeneously across many of the socio-demographic variables. This was an unanticipated finding among the study sample and showed that the socio-demographics for Indian takeaway food consumption were conflicting to those for other types of takeaway food. Physical access and availability of unhealthy activities (takeaway food, smoking, and alcohol) was more prevalent in individuals with a lower socio-economic status, suggesting a strong socio-economic gradient and a clustering of harmful behaviours.

The study of takeaway food in relation to wider dietary patterns in the current study built on the preliminary data describing the frequency of takeaway food consumption, as seen in previous studies (Miura et al., 2012; Adams et al., 2015). Four main dietary patterns produced from the data included a Western dietary pattern, Prudent dietary pattern, Takeaway Food dietary pattern and a Meat and Dairy dietary pattern. Three of the four dietary patterns were characterised by a number of foods that have been associated with poor health outcomes. The Western dietary pattern was positively ($p < 0.005$) associated with younger age, more children in the household, poorer health status, lower education, and single marital status. The Prudent dietary pattern was positively ($p < 0.05$) associated with female gender, older age, higher education, increased alcohol consumption, increased moderate/vigorous exercise, consumption of supplements, non-white ethnicities, and sedentary work. The Takeaway Food dietary pattern was positively ($p < 0.005$) associated with increased adults in the household, poorer health status, smoking, increased alcohol consumption, increased physical daily occupational activity and moderate/vigorous exercise. Finally, the Meat and Dairy pattern was positively ($p < 0.005$) associated with older age, more children in the household, poorer health status, smoking, increased BMI, lower education and increased daily occupational activity. The present study offers valuable insight into the behavioural and societal factors that may contribute to takeaway food consumption and wider dietary patterns in the UK adult population.

Chapter 6
Study 3

6 Metabolite fingerprints in urine associated with takeaway food consumption and wider dietary patterns

Abstract

Background: Accurate measurement of habitual food consumption, particularly energy-dense nutrient-poor foods using self-reported dietary assessment tools can be subject to participant bias. Recent research has demonstrated that metabolites derived from individual foods present in urine samples provide biomarkers of dietary exposure, the measurement of which could improve the limitations of traditional dietary assessment methodologies. Several reports have described the analysis, in human biofluids, of specific metabolites known to be derived from foods of high public health significance, but very few validated biomarkers for takeaway or fast foods. ***Aim of Study 3:*** Investigate the dietary patterns of adult takeaway food consumers, identify metabolite fingerprints and discriminatory biomarkers linked to the consumption of specific foods from spot urine samples, to help validate dietary patterns from self-reported dietary surveys. ***Methods:*** One-hundred and fifty-one participants in Merseyside completed a modified version of the EPIC-Norfolk FFQ and 3 x 24-hour 'Multiple Pass' dietary recalls (24 h MPR). Principal component analysis (PCA) was used to analyse dietary patterns from both dietary assessment tools. Additionally, participants collected first morning void spot urine samples after each 24 h MPR. Metabolite fingerprints were created from urine samples using flow infusion electrospray (FIE) ionisation high resolution (HR) mass spectrometry (MS) and analysed with machine learning data techniques, including Random forest. Ultra High Performance Liquid Chromatography-High Resolution MS (UHPLC-HRMS) and Tandem Mass Spectrometry (MSn) identified the structures of putative biomarkers. ***Results:*** PCA produced four dietary patterns from the FFQ data; 1) Western, 2) Snacks, 3) Healthier, and 4) Takeaway and Meat, and five patterns from the 3 x 24 h MPR; 1) Western, 2) Cosmopolitan, 3) Takeaway, 4) Prudent, and 5) Sweet. FIE-HRMS coupled with machine learning established metabolite fingerprint patterns for consumers of fast food, Chinese, Indian, English and kebab takeaway, showing good biomarker potential and validating self-reported dietary intake. Urinary metabolites associated with habitual exposure to specific foods were identified including poultry, coffee, alcohol etc. ***Conclusion***: Metabolite fingerprints correlated with patterns derived from self-reported dietary surveys and highly discriminatory urinary biomarkers were identified related to consumption of certain foods. These urine biomarkers provided further validation of previously known biomarkers, which are being integrated into a diagnostic population screening method in order to objectively measure food exposure.

6.1 Chapter outline

Work presented in this chapter is from a collaborative study undertaken with the High Resolution Metabolomics Laboratory in the Institute of Biology, Environmental and Rural Sciences (IBERS) at Aberystwyth University (AU). The aim of this collaboration was to determine if metabolite fingerprints from spot urine samples were correlated with dietary patterns derived from self-reported dietary surveys, particularly in the context of consumption of takeaway foods. The team, led by Prof John Draper, are currently developing an extensive biomarker panel covering a range of commonly consumed foods in the UK, including red meat, poultry, wholegrain and cruciferous vegetables from a simple spot urine sample collected at home. However, the current gap in the literature for the acceptability of a home urine collection-kit and biomarkers from energy-dense nutrient-poor foods justified the investigation of takeaway foods.

Two traditional and subjective, dietary assessment tools (FFQ and 3 x 24 h MPR respectively) were included in this study to create several layers of analysis for validation. A sub-sample of participants from the Eating Habits Survey (EHS) participated in the current study, thus previously collected data in the EHS (FFQ dietary assessment) were analysed to explore the dietary patterns of this sub-sample. Three 24 h MPR were then conducted on the sub-sample to measure consumption of takeaway foods as well as define wider dietary patterns. Participants involved in the 3 x 24 h MPR provided spot urine samples each morning at home using a bespoke urine collection-kit, following the 3 x 24 h MPR, for metabolomic analysis at IBERS using flow infusion electrospray (FIE) ionisation high resolution (HR) mass spectrometry (MS). Urinary metabolite fingerprints were then subsequently compared with the consumption of takeaway foods, habitual foods and wider dietary patterns.

6.2 AIMS AND OBJECTIVES

Aim: To identify the dietary patterns of a sub-sample of takeaway food consumers and to add a model for objective classification of dietary exposure based on urinary metabolite fingerprint data.

Objectives:

1. Distinguish a sub-sample of suitable participants for Study 3 and identify separate groups of takeaway food consumers.
2. Classify dietary patterns of study sample (from the EHS and 3 x 24 h MPR) and test the validity of the EHS as a dietary assessment tool.
3. Develop metabolite fingerprints of high and low takeaway food consumers.
4. Determine if the metabolite fingerprints of urine samples validate participant dietary intake from the 3 x 24 h MPR and validate known food metabolites or novel biomarkers.
5. Determine if metabolite fingerprints reflect habitual dietary patterns and components.
6. Evaluate the acceptability of the bespoke home urine collection-kit.

6.3 METHODS

6.3.1 STUDY POPULATION

To meet an initial 200 sub-sample target set for Study 3 (by IBERS, AU), all eligible habitual takeaway food consumers identified in the EHS (FFQ in Study 2) were sent an e-mail and/or text message (Appendix 9.27), including a participant information sheet (Appendix 9.28), inviting them to participate in the final study. Participants had one week to decide to take part or not. Recruitment took place from April to October 2017, during this period 190 participants (adults aged 18–64 years) provided preliminarily verbal consent and received a bespoke urine sampling kit through the post. The kit included a participant information sheet, written informed consent form (Appendix 9.29), photographic instructions sheet for urine collection (Appendix 9.30) and a 3-day food record (Appendix 9.31). After drop-out (n = 35) and exclusions (n = 4) due to missing data 151 participants (79.5%), 34 males and 117 females, had provided written informed consent, completed 3 x 24 h MPR and provided 3 days spot urine samples. It was initially decided to collect multiple urine samples from 200 participants to enable the investigation of correlations between foods consumed over 3 days and the metabolites in urine samples at a population level, however 151 was deemed enough. This target also met the minimum of 150 participants needed to derive dietary patterns through principal component analysis (PCA) (Pallant, 2013). Finally, each participant received a debrief on close of the project.

6.3.2 Collection of data from FFQ and 3 x 24 h MPR

A data export from the EHS (Study 2) for the 151 participants in this study helped to characterise the participants, explore their dietary patterns, validate data from 3 x 24 h MPR, and aided the interpretation of inter- and intra-individual differences in metabolite fingerprints. Participants completed a 3-day 24 h food record (Appendix 9.31) for all food and drink consumed on their chosen non-consecutive days Monday–Sunday (2 weekdays and 1 weekend day) to assess food consumption. They also collected a urine sample in their own home using the bespoke urine sampling kit the morning following each 24 h food record. On the day of urine collection, participants completed a telephone interview 24 h MPR; in accordance to the USDA 5 step multiple pass method (Fig. 6.1). Participants were able to refer back to their food record to help with memory and estimated portion sizes using household measures and grams weights.

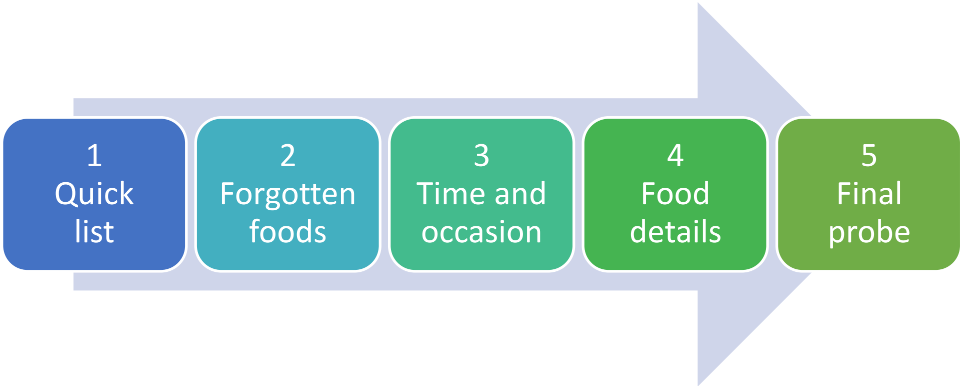


Figure 6-1 The 5 step 24 hour 'Multiple Pass' dietary recall method

6.3.3 HUMAN TISSUE ACT AND SAMPLE STORAGE

In April 2016, at the crucial development stage of Study 3, Prof Khalid Rahman from the School of Pharmacy and Biomolecular Sciences at LJMU provided additional expertise. This included discussions relating to the logistics of collecting and storing human tissue in compliance with Human Tissue Authority (HTA) regulations (Human Tissue Authority, 2018) and for the appropriate measures to be in place prior to the transfer of human tissue to an external institution (AU), specifically the materials transfer agreement (MTA). HTA training was completed in May 2016 and Prof Rahman was added to the project for his experience of working with human tissue. Following, Alan Simm, the Technical Research Manager for the Faculty of Science, liaised to organise freezer storage space for the duration of data collection and requested a protocol for the study and risk assessment for samples (Appendix 9.32), along with a standard operating procedure (Fig. 6.2). Furthermore, John Hall, the Laboratory Manager & HTA Tissue Coordinator of the life sciences laboratory delivered laboratory health and safety training. Agreement of the MTA occurred on 7th December 2016 (Appendix 9.26).

In relation to the management of urine samples, processing and storage from April to October 2017, good working relationships with staff receiving samples at the Goods Reception was essential. Prior to the collection of samples, staff received a protocol explaining the correct storage of samples in a temporary −20°C freezer (Appendix 9.33). Batches of samples, collected from the −20°C freezers on a weekly basis, were prepared for storage in the −80°C freezers in the Life Science laboratory. The Pro-curo database (Pro-curo Software Ltd. West Sussex, UK) stored sample information and codes to increase sample traceability and identification of human tissue samples at any point during the project lifetime. All written informed consent forms were received separately from urine samples and stored in designated HTA lockable filing cabinets, located in the Life Sciences building, to meet HTA requirements (Fig. 6.2). Samples were then temporarily stored in −80°C designated HTA freezers in preparation for transfer to IBERS, AU. An initial batch of samples (n = 237) for 40 participants (6 samples per participant) were packaged on dry ice and transferred to AU under MTA for analysis in the IBERS laboratories in August 2017 (Fig. 6.2). Researchers at IBERS rendered samples acellular within 7 days of arrival in compliance with HTA regulations. On completion of sample collection, the final batch of samples (n = 665) for the remaining 111 participants (6 samples per participant) were sent following the same protocol to AU in February 2018.

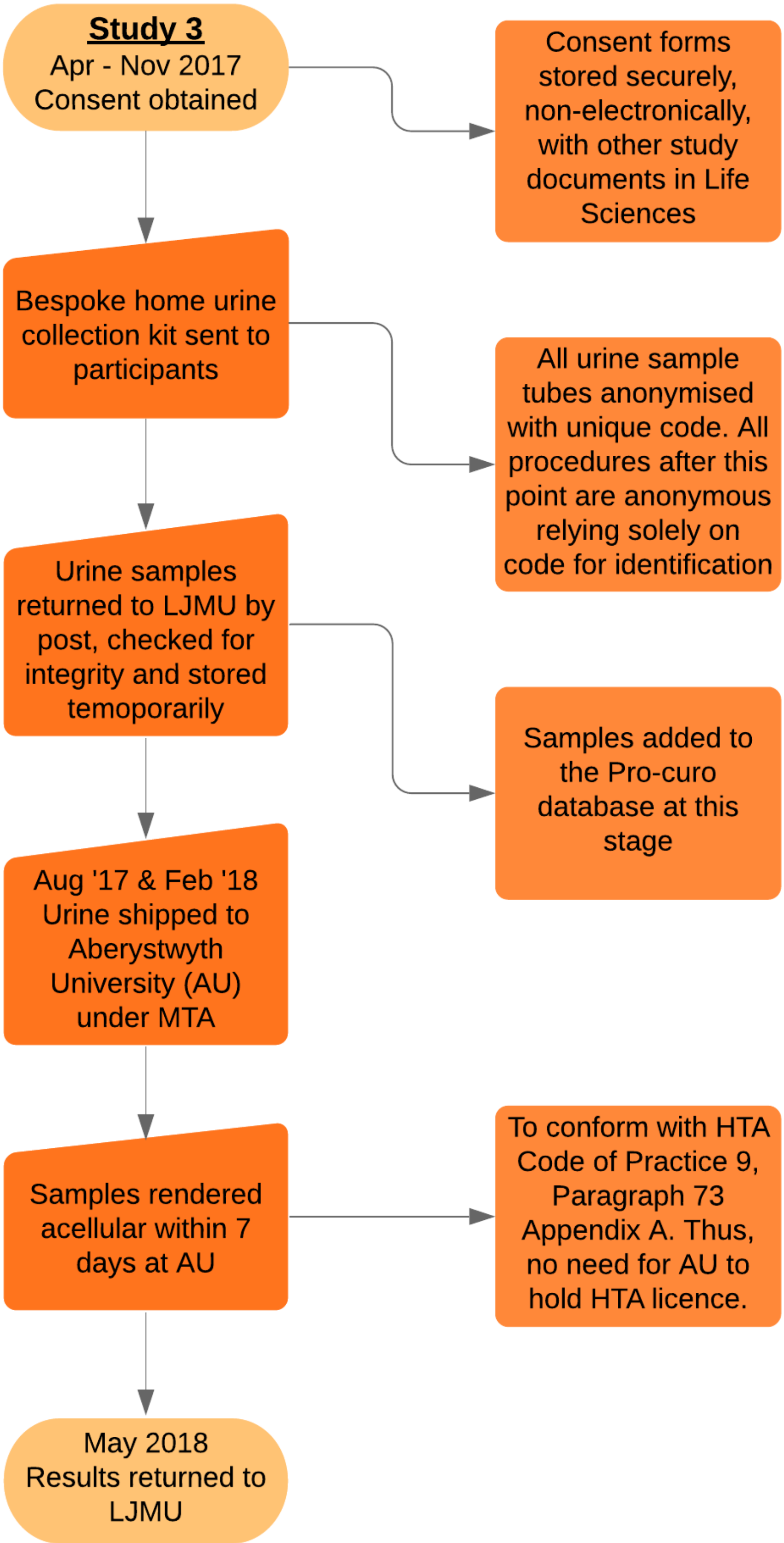


Figure 6-2 Flowchart of urine sampling management protocol

6.3.4 Bespoke home urine collection

Bespoke urine sampling kits (Fig. 6.3) (Wilson et al., 2016) were sent out to consenting participants along with a 3-day food record, an instructions sheet and a pre-paid Royal Mail Safebox™ (approved for the sending of samples and specimens, fully compliant with UN3373 requirements) for safe return of samples to LJMU premises. Data collection occurred between April and October 2017. Collection of spot morning void urine samples was favourably selected against 24 hour urine collection due to the reduced impact on normal daily activities and logistical costs (Fave et al., 2011). A 24 hour urine collection can provide robust and accurate quantification of analytes however, its collection imposes a significant burden on study participants due to its adverse impact on normal daily activities. There is also a significant risk that measurements are misreported due to incorrect collections during 24 hour urine collection. Previous research has shown that spot urine sample collections can adequately substitute for 24 hour urine samples for biomarker discovery and habitual dietary exposure measurements (Beckmann et al., 2013). Therefore, participants collected spot first morning void urine samples after each 24 h MPR. Two 4ml urine sample tubes were filled each morning to correlate with all food and drink consumed during the previous 24 h period, contributing to a total of six samples tubes each (3 samples for analysis and 3 for contingency). Participants received reminder texts during the study to check for compliance, prompt their collection of urine samples and organise a convenient time to contact by telephone for the 24 h MPR. Urine samples were stored in the participant's fridge and once all 3 days were complete, were posted back in pre-paid Safeboxes™ to LJMU premises.

Figure 6-3 Urine collection-kit used in the study

6.3.5 URINE COLLECTION-KIT ACCEPTABILITY

At the end of the experimental period, eligible participants (individuals that had successfully completed the 3 x 24 h MPR and urine sample collection) received an e-mail invitation (Appendix 9.34) with participant information sheet (Appendix 9.35), and link to complete a short acceptability questionnaire on Online Surveys (Jisc, UK) (Appendix 9.36). The purpose of the questionnaire was to evaluate the usability and ease of use of the bespoke home urine collection and postal kit with the aim to improve the kit for future research purposes. Of the 155 participants invited to participate, 140 participated (90% response rate) in the acceptability questionnaire.

6.3.6 STATISTICAL ANALYSIS OF FFQ, 3 X 24 H MPR AND ACCEPTABILITY QUESTIONNAIRE

Statistical analysis of data was performed using IBM SPSS® 24.0 (SPSS Inc., Chicago, Illinois, USA) to explain the socio-demographic and lifestyle characteristics of the population, evaluate the acceptability of the urine sampling kits, and to investigate the dietary patterns from two dietary assessment techniques (FFQ and 3 x 24 h MPR). Food and drink portions in the 3 x 24 h MPR were converted into servings for the three days and transposed into an excel workbook. Foods were aggregated into 33 food groups (Table 6.1) in line with commonly consumed foods, foods of key interest, and foods where biomarkers are available (for quantification purposes). Food groups were then imported to SPSS for dietary pattern analysis (PCA as described previously (Study 1, Chapter 4.3.3) to produce primary components, which accounted for variation in dietary intake. On inspection of the correlation matrix, food groups that had values < 0.25 were removed from final PCA analysis due to having low/no influence on the patterns. For PCA on the 3 x 24 h MPR, this resulted in other potato, savoury sauces, fish and shellfish, alcohol, tea, Indian takeaway, English takeaway and kebab takeaway being omitted due to minimal influence on dietary patterns, suggesting that low quantitates were consumed by participants for the 3 days (Table 6.1). Whereas, for PCA on the FFQ data, tea and coffee, and alcohol were omitted due to low influence on the dietary patterns.

Table 6-1 Food grouping used in the dietary pattern analysis of the 3 x 24 h MPR cohort

Food Group	Definition and content
1. Vegetables	Carrots, spinach, broccoli, brussel sprouts, cabbage, peas, green beans, courgettes, cauliflower, parsnips, leeks, onions, garlic, mushrooms, sweet peppers, beansprouts, green salad,

		mixed vegetables, watercress, tomatoes, sweetcorn, beetroot, coleslaw
2.	**Fruit**	Apples, pears, oranges, grapefruit, bananas, grapes, melon, peaches/plums, strawberries/raspberries/other berries, avocado, tinned fruit, dried fruit
3.	**Nuts and seeds**	Nuts, seeds, nut butters and 1% from nut milk
4.	**Legumes**	Beans, pulses, soy products, 1% from soya milk
5.	**Wholegrains/meal**	Brown bread, wholemeal bread, all bran, wholegrain cereals, porridge, brown rice, wholemeal pasta
6.	**Other grains**	White bread, crackers, pitta/naan, garlic bread, sugary cereal, non-sugary cereal, pasta, tinned pasta, super noodles, pizza, white rice
7.	**Biscuits**	Sweet plain biscuits, sweet chocolate biscuits, cookies
8.	**Buns, cakes, pastries, fruit pies**	Cakes, scones, sweet buns/pastries, fruit pies, Yorkshire puddings
9.	**High salt snacks**	Crisps, salty popcorn
10.	**Fried potato**	Chips, wedges, roast potatoes, fries
11.	**Other Potato**	Boiled potatoes, baked potato
12.	**Sugar**	Sugar, honey, syrup
13.	**Cocoa**	Chocolate, hot chocolate
14.	**Sweet condiments**	Ketchup, brown sauce, BBQ sauce, relishes, jam, chocolate spread
15.	**Savoury sauces**	Gravy, stock, marmite, soy sauce
16.	**Red and processed meat**	Beef, burgers, pork, lamb, bacon, ham, corned beef, sausages, savoury Pies, offal/pate, lasagne
17.	**Poultry**	Chicken, turkey
18.	**Fish and shellfish**	Fish, fish Fingers, white fish, oily fish, shellfish
19.	**Egg**	Eggs, quiche, 2% from mayonnaise
20.	**Dairy**	Single/sour cream, double or clotted cream, low fat yoghurt, full fat yoghurt, dairy desserts, cheese, cottage cheese, butter, milk pudding, ice cream, coffee whitener
21.	**Oils and dairy free spreads**	Block margarine/stork, polyunsaturated margarine/flora, other soft margarine/clover, low fat spread/gold, low calorie salad cream/mayonnaise, regular salad cream/mayonnaise, French dressing, other salad dressings
22.	**Alcohol**	All alcoholic drinks
23.	**Tea**	Black tea, decaffeinated black tea
24.	**Coffee**	Coffee, decaffeinated coffee
25.	**Sugar-sweetened beverages**	Sugar-sweetened carbonated soft drinks, squash/cordial, sweeteners

26. Sugar-free beverages	Sugar-free carbonated soft drinks, sugar-free squash/cordial
27. Fruit juice	Pure fruit juice
28. Fast Food	Beef burger, chicken burger, fried chicken, fish burger, veggie burger, mozzarella sticks, fries, McDonald's breakfast, Subway, sauces/dips
29. Chinese takeaway	Chicken wings, spare ribs, crispy duck, spring rolls, prawn crackers, siu mai, skewered chicken, soup, salt and pepper chicken, peking dish, lemon/orange/plum sauce dish, satay dish, sweet and sour, curry, chop suey, kung po, foo yung, chow mein, beef dish, chicken or Duck Veg, king prawn, fried rice, salt and pepper chips, noodles, boiled rice, chinese sauces
30. Indian takeaway	Poppadums, samosa, tikka starter, tandoori, korma, tikka masala, rogan josh, bhuna, biryani, balti, sagwala, tikka jalfraizi, dupiaza, medium curry, achari, vindaloo, pathia, pasanda, special, butter chicken, saag, dal, naan, paratha, pilau, fried rice, boiled rice
31. English takeaway	Fish, scampi, sausage, omelette, roast chicken, chips, sauces
32. Pizza takeaway	Cheese, meat, seafood, vegetarian, garlic bread
33. Kebab takeaway	Donner, chicken, sheesh

NB: Food groups removed from final PCA analysis highlighted in red

Foods from the FFQ data (previously collected during Study 2) were aggregated into similar food groups to those from the 3 x 24 h MPR (Table 6.2); this was to ensure comparability during analyses of FFQ and 24 h MPR dietary patterns. PCA determined dietary patterns in SPSS as described previously (Study 1, Chapter 4.4.4) on 31 food groups from the FFQ data for 151 participants. The components (dietary patterns) were labelled based on an interpretation of the data and of the earlier literature.

Table 6-2 Food grouping used in the dietary pattern analysis of the FFQ sub-sample cohort

Food Group	Definition and content
1. Vegetables	Carrots, spinach, broccoli, brussel sprouts, cabbage, peas, green beans, courgettes, cauliflower, parsnips, leeks, onions, garlic, mushrooms, sweet peppers, beansprouts, green salad, mixed vegetables, watercress, tomatoes, sweetcorn, beetroot, coleslaw
2. Fruit	Apples, pears, oranges, grapefruit, bananas, grapes, melon, peaches/plums, strawberries, avocado, tinned fruit, dried fruit
3. Nuts	Peanuts, peanut butter
4. Legumes	Baked beans, dried lentils, tofu
5. Wholegrains/meal	Brown bread, wholemeal bread, all bran, wholegrain cereals, porridge, brown rice, wholemeal pasta
6. Refined grains	White bread, crackers, pitta/naan, garlic bread, sugary cereal, non-sugary cereal, pasta, tinned pasta, super noodles, pizza, white rice
7. Buns, Cakes, Pastries, fruit pies	Cakes, scones, sweet buns/pastries, fruit pies, Yorkshire puddings
8. Biscuits	Sweet biscuits (chocolate), sweet biscuits (plain)
9. Chocolate	Chocolate
10. Fried potato	Chips, roast potatoes
11. Other Potato	Boiled potato/baked potato, potato salad
12. Tea and Coffee	Tea, coffee
13. Sweets and Sugar	Sweets, sugar
14. Sweet Sauces and condiments	Ketchup, relishes, jam/syrup/honey, chocolate spread
15. Savoury Sauces	Vegetable soup, meat soup, gravy/cheese sauce/white sauce, marmite, dips, tomato sauces
16. High Salt Snacks	Crisps
17. Red and processed meat	Beef, burgers, pork, lamb, bacon, ham, corned beef, sausages, savoury Pies, offal/pate, lasagne
18. Poultry	Chicken, turkey
19. Fish and shellfish	Fish, fish Fingers, white fish, oily fish, shellfish
20. Egg	Eggs, quiche
21. Dairy	Single/sour cream, double or clotted cream, low fat yoghurt, full fat yoghurt, dairy desserts, cheese, cottage cheese, butter, milk pudding, ice cream, coffee whitener

22. Fats and oils (not inc butter)	Block margarine/stork, polyunsaturated margarine/flora, other soft margarine/clover, low fat spread/gold, low calorie salad cream/mayonnaise, regular salad cream/mayonnaise, French dressing, other salad dressings
23. Alcohol	Wine, beer/larger, liqueurs, spirits
24. Sugar-sweetened beverages	Cocoa, Horlicks, Sugar-sweetened carbonated soft drinks, pure fruit juice, squash/cordial
25. Sugar-free beverages	Sugar-free carbonated soft drinks
26. Fast Food	Beef burger, chicken burger, fried chicken, fish burger, veggie burger, mozzarella sticks, fries, McDonald's breakfast, Subway, sauces and dips
27. Chinese takeaway	Chicken wings, spare ribs, crispy duck, spring rolls, prawn crackers, siu mai, skewered chicken, soup, salt and pepper chicken, peking dish, lemon/orange/plum sauce dish, satay dish, sweet and sour, curry, chop suey, kung po, foo yung, chow mein, beef dish, chicken or Duck Veg, king prawn, fried rice, salt and pepper chips, noodles, boiled rice, chinese sauces
28. Indian takeaway	Poppadums, samosa, tikka starter, tandoori, korma, tikka masala, rogan josh, bhuna, biryani, balti, sagwala, tikka jalfraizi, dupiaza, medium curry, achari, vindaloo, pathia, pasanda, special, butter chicken, saag, dal, naan, paratha, pilau, fried rice, boiled rice
29. English takeaway	Fish, scampi, sausage, omelette, roast chicken, chips, sauces
30. Pizza takeaway	Cheese, meat, seafood, vegetarian, garlic bread
31. Kebab takeaway	Donner, chicken, sheesh

NB: Food groups removed from final PCA analysis highlighted in red

6.3.7 URINE SAMPLE PREPARATION (EXTRACTION) AND ADJUSTMENT

Samples (n = 453) were placed into racks and defrosted from a −80°C freezer overnight in a 4°C fridge. Once defrosted, samples were centrifuged (600 xg for 5 mins at 4°C to separate the liquid in the urine from any solid components, as recommended by Becton Dickinson Biosciences, UK). The samples were then placed on ice and aliquots of thawed urine (1000 µL) was transferred into labelled

2 mL Eppendorf tubes (220 µL plus 500 µL for extraction). The remaining samples were returned to a −20°C freezer. An OPTI Hand Held Refractometer (Bellingham Stanley™ Brix 54 Model) was calibrated with de-ionised water (dH_2O) and dried with tissue according to the manufacturer's instruction. Following, 220 µL of sample was transferred onto the refractometer dish, the specific gravity (SG) value was recorded in triplicate and temperature was noted. The refractometer was rinsed with dH_2O between samples and dried with tissue. Specific gravity values were used to calculate average values before being used to calculate urine_V_m and H2O_V_m for each sample. Based on these figures, aliquots of the required amounts of urine (from vortexed and centrifuged 2ml Eppendorf tubes) and dH2O were transferred into new 2ml Eppendorf tubes on ice to make up 500 µL for extraction. In addition, 500 µL aliquots of pre-chilled (−20°C) extraction methanol (Primer-Trace analysis grade) was added to each adjusted sample and vortexed before being refrigerated at 4°C overnight. To pool urine for each individual, aliquots from each sample (20 µL) was transferred into new 2ml Eppendorf tubes (n = 151). 20 µL of extracted sample was transferred to a glass HPLC vial containing a 200 µL flat bottom micro insert (Chromacol). All samples were diluted with 80 µL of pre-chilled methanol/water (MeOH:H2O, 70:30) directly in the vial for flow infusion. A master mix was also created by adding 20 µL of each extracted sample into a 5ml Eppendorf tube.

6.3.8 Flow Infusion-High Resolution Fingerprinting (FIE-HRMS)

All samples (n = 151) were analysed using high resolution (HR) flow infusion electro-spray (FIE) ionisation mass spectrometry (MS) as described previously

(Beckmann et al., 2008). Mass spectra were acquired on an Exactive Orbitrap (ThermoFinnigan, San Jose, CA) mass spectrometer coupled to an Accela (ThermoFinnigan, San Jose, CA) ultra-performance liquid chromatography system. A 20 μL of sample was injected and delivered to the ESI source via a flow solvent (mobile phase) or pre-mixed HPLC grade MeOH (Fisher Scientific) and ultra- pure H2O (18.2 Ω) at a ratio of 70:30. The flow rate was 200 μLmin-1 for the first 1.5 minutes, and 600 μLmin-1 for the remainder of the method. The total method time was 3.0 minutes. Positive and negative ionisation modes were acquired simultaneously. During the electrospray process, ionisation is accomplished by the loss or gain of a proton, or other adducts charged through the formation of adducts such as Cl- ions in negative ion mode or commonly K or Na in positive ion mode) to allow flow and detection. One scan event acquired each mass spectrum, 55.000–1200 m/z for both positive and negative mode. The scan rate was 1.0 Hz. Mass resolution was 100,000, with an automatic gain control (AGC) 5 x 105 and maximum injection time of 250 ms, for both ionisation modes.

6.3.9 Statistical analysis of FIE-HRMS data

Analysis of the fingerprint data was carried out by researchers at IBERS, AU and a brief description of their methods are described below. Raw files were exported to CDF-files, mass aligned and centroided in MATLAB (V8.2.0, The MathWorks) maintaining highest mass accuracy. Mass spectra around the apex of the infusion peak were combined in a single intensity matrix (runs x *m/z*) for each ion mode, and binning the *m/z* and intensity values at 0.01 amu intervals. Data from intensity matrix was log-transformed before further statistical analysis. Data mining was carried out using the FIEmspro workflow previously validated (Enot et al., 2008) and accessible online (Beckmann et al., 2007). Wherever possible, a minimum of 12 participants was used for each target food group (e.g. Chinese) or dietary pattern (e.g. Western) to study metabolite fingerprints; previous research has suggested 8–20 participants to target urinary signals of specific foods (Lloyd et al., 2013). Principal component analysis (PCA) reduced data dimensionality and was followed by principal component–linear discriminant analysis (PC-LDA). Plots of the first two PC-Discriminant Functions (PC-DFs) allowed visualization of the goodness of class separation as quantified by Tw values (Eigenvalues). Discrimination was considered adequate for Tw values > 2.0 and poor for Tw values < 1.0 (Enot et al., 2008). Participants were classified as very high consumers (> 10), high consumers (> 4.0) and low consumers (< 2.0) for consumption of specific takeaway foods in three days, and non-consumers. Those that did not fit into these groups were omitted from the analysis.

PC-LDA was performed using both positive and negative ionisation modes of data because each mode can produce different ionisation adducts and signals. In previous literature, the negative ionisation mode has been favoured for showing more signals, and therefore more data rich, than the positive mode (Lloyd et al., 2011a). This is partly because biotransformation products (glucuronides and sulphates) of urinary food biomarkers occur in the negative ionisation mode, which are relatively easy to determine. Nonetheless, other ionisation adducts and signals can be better determined using the positive mode. The urinary chemistry for high and low consumers of each takeaway sub-group (fast food, Indian, Chinese, English, pizza and kebab) and controls (non-consumers of all take-away sub-groups) were compared to identify known and novel markers associated with takeaway consumption.

Supervised Random Forest (RF) classification was implemented using the randomForest package in R (R Core Team, 2013). For all RF models, the number of trees (*ntree)* used was 1000 and the number of variables considered at each internal node (mtry) was the square root of the total number of variables. Accuracy, margins of classification and area under the Receiver Operator Characteristic (ROC) curve (AUC) were used to evaluate the performance of classification models, as previously described (Enot et al., 2008). Models were deemed adequate overall if RF margins > 0.2 and AUC values > 0.8, which have been implemented previously (Enot et al., 2008; Lloyd et al., 2016).

6.3.10 Targeted accurate mass analysis and annotation of FIE-HRMS signals/MZedDB, correlation and identification of isotopes

For metabolite signal annotation, accurate *m/z* values were extracted from the un-binned matrix to enable direct identification of metabolites at 1-5 ppm directly in the first pass profile. These were queried using MZedDB, an interactive accurate mass annotation tool used to directly annotate signals by means of neutral loss and/or adduct formation rules (Draper et al., 2009). Pearson correlation coefficients between selected variables were calculated. Variables with correlation coefficients > 0.7 were considered to belong to a cluster indicative of different ionisation or potential biotransformation/breakdown products of a parent food-derived metabolite. Visual analysis of the spectra allows identification of isotopes of biomarkers of interest allowing further confirmation of correct identification of signals. Data from the 24 h MPR was analysed and used in this analysis for validation purposes.

6.4 Results

6.4.1 Participant characteristics

The total study population was 151 (117 female, 34 male) adult participants from Merseyside (Table 6.3). Four exclusions had been made due to missing data (either urine samples or 24 h MPR data). A number of participants failed to report their height and weight, waist circumference and alcohol consumption in the EHS. However, they were not removed for the analysis as this missing data was not considered the primary focus of the study. The study population's socio-demographic (Table 6.3) and lifestyle (Table 6.4) characteristics is described below.

Table 6-3 Study population socio-demographic characteristics 2017 (n = 151)

Variable	Level	*n*	%
Age group (years)	18 – 25	25	16.6
	26 – 35	52	34.4
	36 – 45	38	25.2
	46 – 55	24	15.9
	56 – 64	12	7.9
Gender	Male	34	22.5
	Female	117	77.5
Ethnicity	White	127	84.1
	Non-White	24	15.9
Marital status	Single	72	47.7
	Married (inc civil partnership)	68	45.0
	Divorced/Separated/Widowed	11	7.3
Living arrangements	On own	20	13.2
	Wife/Husband/Partner	88	58.3

	Parents	13	8.6
	Friends	9	6.0
	Children	20	13.2
	Other	1	0.7
Adults in household (18+)	1	32	21.2
	2	83	55.0
	3	24	15.9
	4 or more	12	7.9
Children in household (U18)	0	75	49.7
	1	31	20.5
	2	33	21.9
	3	11	7.3
	4 or more	1	0.7
Educational achievement	GCSE/O-Level	18	11.9
	A-Level or Equivalent	29	19.2
	Degree Level	66	43.7
	Postgraduate Qualification	38	25.2
Employment status	Employed/self-employed	96	63.6
	Not in employment	17	11.3
	Full Time Student	28	18.5
	Retired	5	3.3
	Other (Carer/ill health)	5	3.3
NS-SEC	Managerial/professional	55	36.4
	Intermediate	30	19.9
	Routine/manual	20	13.2
	Student/Unemployed/Retired	46	30.5

NB: n = number of participants. NS-SEC = National Statistics Socio-economic Classifications. U18 = Under 18 years of age.

Table 6-4 Study population self-reported lifestyle and anthropometric characteristics 2017 (n = 151)

Variable	**Level**	***n***	**%**	**Mean (SD)**
BMI (kg/m²)		148		26.8 (6.2)
WHtR (cm)		95		0.5 (0.1)
Alcohol consumption (drinks/week)		147		5.2 (6.4)

Smoking Status	Smoker	21	13.9	
	Non-Smoker	130	86.1	
Daily occupational activity	Sedentary	68	45.5	
	Standing	68	44.2	
	Manual	14	9.7	
	Heavy	1	.6	
Leisure activity (minutes/day)	LPA	151		72.5 (62.9)
	MVPA	151		52.0 (56.2)
Supplement use	Yes	76	50.3	
	No	73	48.3	
	Not Sure	2	1.3	
Health status	Very Good	37	24.5	
	Good	43	28.5	
	Neither Good nor Poor	50	33.1	
	Poor	19	12.6	
	Very Poor	2	1.3	
Long-term illness	Yes	48	31.8	
	No	103	68.2	

NB: *n* = number of participants. M = Mean. SD = Standard deviation. LPA = Low physical activity. MVPA = Moderate and vigorous physical activity.

6.4.2 ACCEPTABILITY OF THE HOME URINE COLLECTION-KIT

The majority (96%) of participants were successful in collecting urine samples using the home urine collection-kit (Fig. 6.4). Only 3% of the participant sample disagreed or strongly disagreed with the statement regarding successful collection.

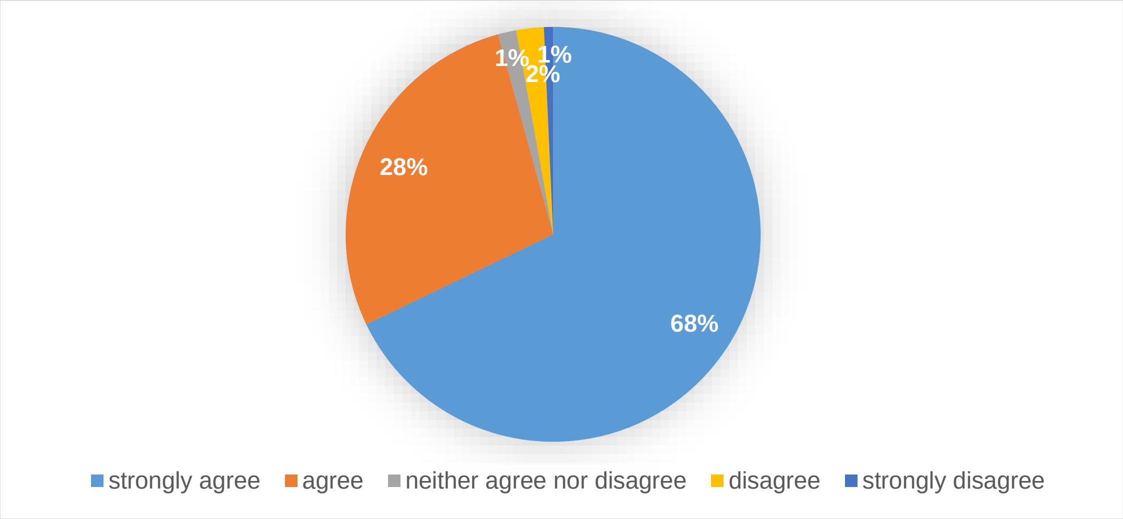


Figure 6-4 I was successful in collecting urine using this method (n = 140)

When asked if participants thought urine sample collection was difficult, the majority (85%) reported that they strongly disagreed or disagreed with the statement. Whereas 15% reported difficulty during urine collection (Fig. 6.5).

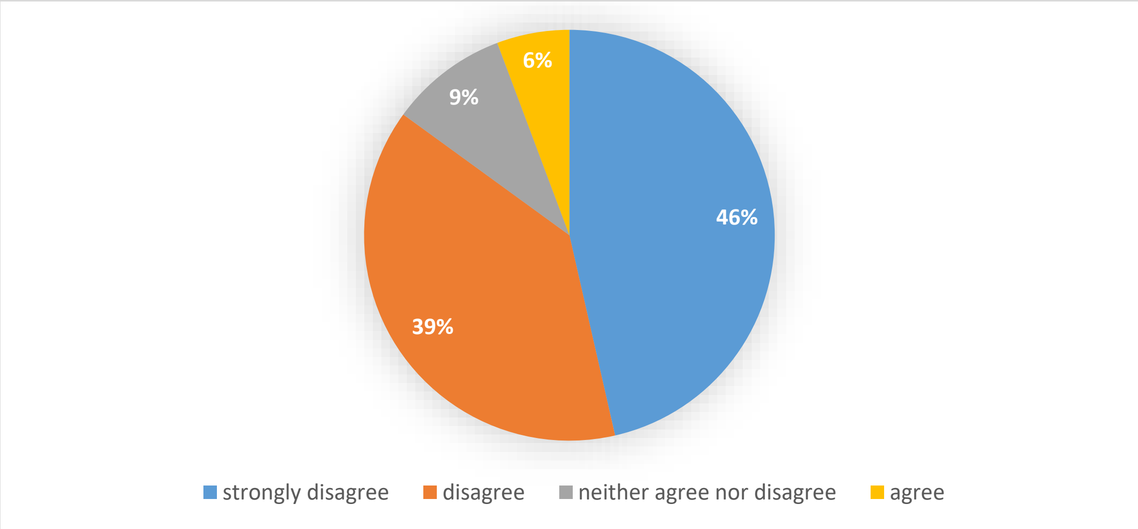


Figure 6-5 In general, I think collecting a urine sample is difficult (n = 140)

A high percentage of participants either strongly agreed (69%) or agreed (26%) with the statement that they were happy to post the urine samples back in a pre-paid box (Fig. 6.6). Only 2% of the participant sample disagreed or strongly disagreed.

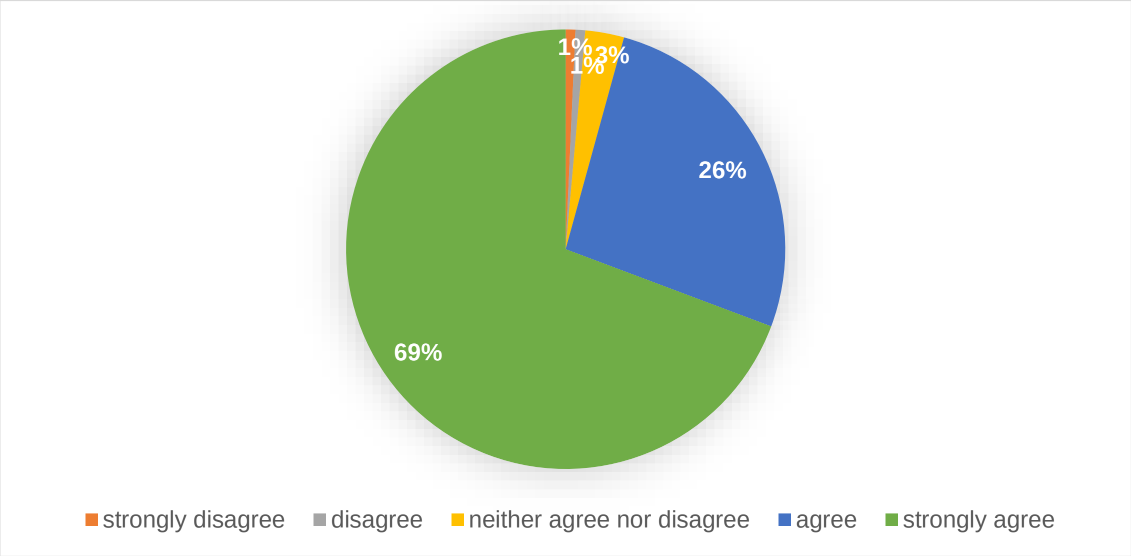


Figure 6-6 I was happy to post urine samples in a pre-paid box (n = 140)

When asked about collecting first morning void urine samples, 87% said they were happy doing so and only 2% said that they were not (Fig. 6.7).

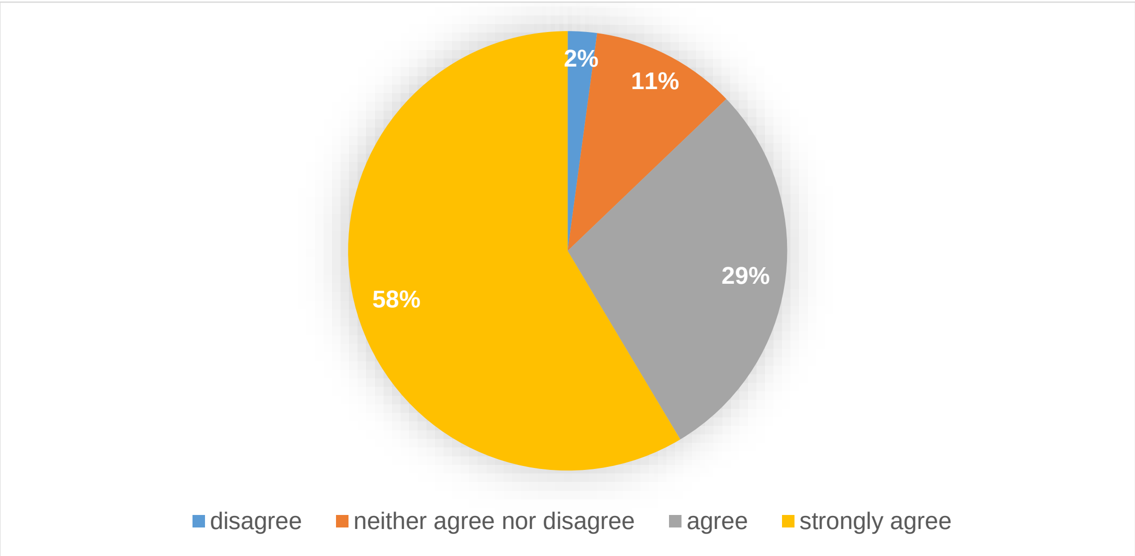


Figure 6-7 I was happy collecting first morning void samples (n = 140)

6.4.3 Dietary patterns from the Eating Habits Survey and 24 h MPR

6.4.3.1 Eating Habits Survey (FFQ)

PCA on 31 food groups was used to analyse habitual and takeaway food intake of 151 participants. The suitability of PCA was assessed prior to analysis. Inspection of the correlation matrix showed that most variables had at least one correlation coefficient greater than 0.25, those that did not have a correlation of > 0.25 were removed from the analysis (alcohol and tea and coffee), leaving a remainder of 29 food groups to re-test. The overall Kaiser-Meyer-Olkin (KMO) measure was 0.70 with individual KMO measures all greater than 0.7, classifications of 'meritorious' to ‘marvelous’ according to Kaiser (Kaiser, 1974). Bartlett's Test of Sphericity (Bartlett, 1954) reached statistical significance ($p < 0.0005$), supporting the factorability of the correlation matrix.

PCA revealed eight components that had eigenvalues > 1, which explained 20.5%, 9.7%, 7.6%, 7.2%, 5.6%, 5.5%, 4.2% and 4.1% of the total variance, respectively. However, a graphical evaluation of the scree plot (Appendix 9.37) showed a clear break after the fourth component, therefore it was decided to retain four components (Cattell, 1966). The four-component solution met the interpretability criterion and this solution explained 45.0% of the total variance.

The first pattern to emerge from the orthogonal (varimax) rotation was described as a ‘Western’ pattern. This was characterised by high positive loadings (> 0.3) for English takeaway (0.769), sweet/sauces/condiments (0.738), legumes (0.676), fast food (0.630), SSB (0.540), chocolate (0.506), fried potato (0.501), other potato (0.391), sweets and sugar (0.382), diet drinks (0.357), biscuits (0.341), and high-salt snacks (0.364) (Table 6.5). There were no negative loadings > −0.3 in this pattern.

The second pattern was a ‘Snacks’ pattern with high positive factor loadings (> 0.3) for dairy (0.789), cakes/pastry/buns (0.707), fats and oils (0.643), other grains (0.600), eggs (0.592), biscuits (0.556), high-salt snacks (0.493), Indian takeaway (0.403), chocolate (0.340), fried potato (0.328), other potato (0.380) and sweets and sugar (0.304) (Table 6.5). There were no negative loadings > −0.3.

For the third pattern there were high positive loadings (> 0.3) for vegetables (0.780), nuts (0.634), wholegrains/meal (0.560), fruit (0.551), fish (0.495), savoury sauces (0.492), legumes (0.456), other potato (0.377), pizza (0.337), cakes/pastry/buns (0.323). There were no negative loadings > −0.3 (Table 6.5). Thus, this pattern could be defined as a ‘Healthier’ pattern.

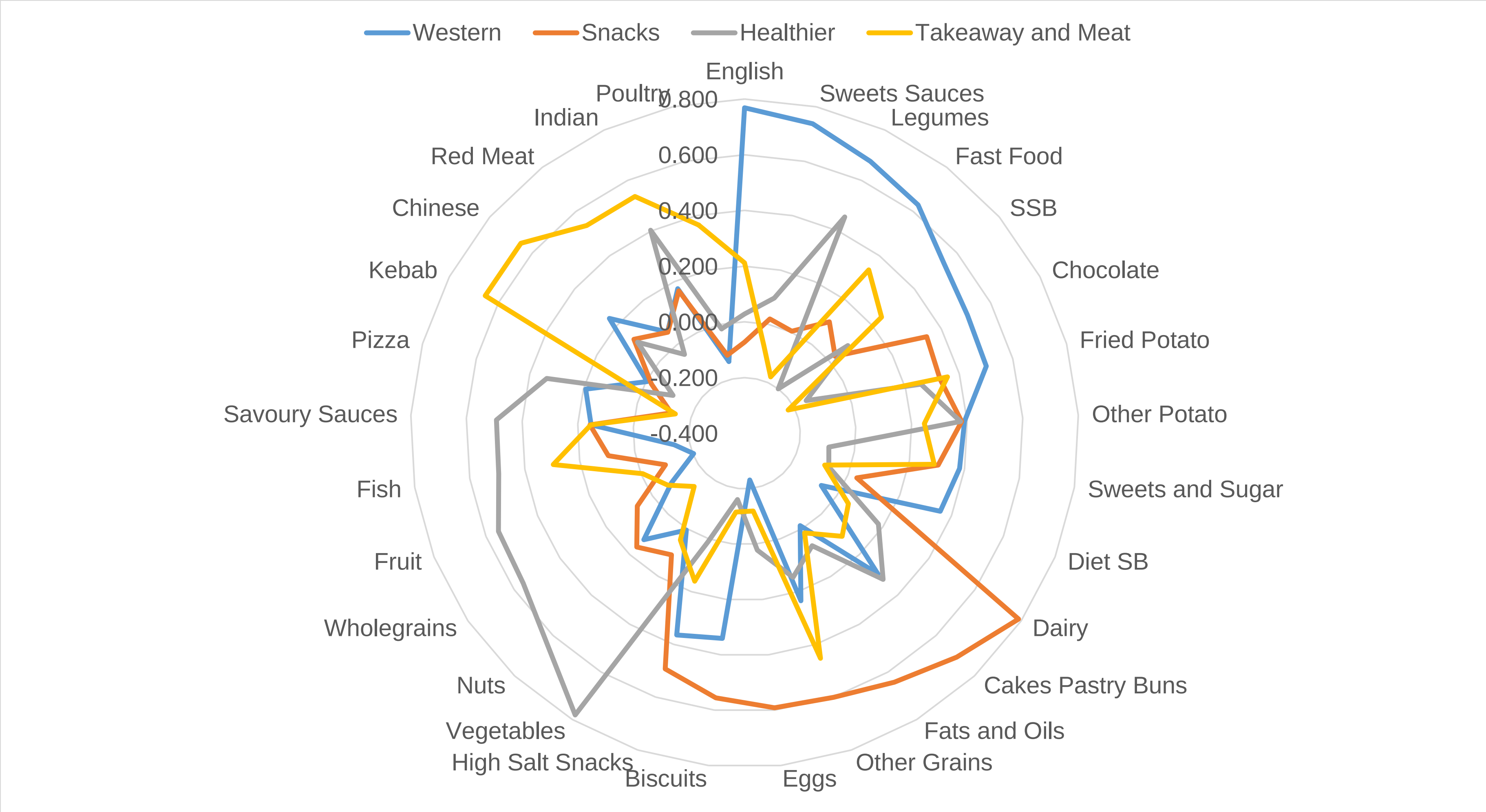
The fourth and final component identified as a ‘Takeaway and Meat’ pattern presented high positive loadings (> 0.3) for kebab (0.654), Chinese (0.654), red meat (0.537), Indian (0.537), other (refined) grains (0.453), poultry (0.366), fast food (0.337) and fried potato (0.356) (Table 6.5). There were no negative loadings > −0.3

The component scores for each dietary pattern from the EHS (FFQ) data were input into a radar graph to provide a visual interpretation of the differences between the four dietary patterns produced through PCA (Fig. 6.8).

Table 6-5 Rotated component loadings derived from PCA with Varimax Rotation on 29 food groups from FFQ results for dietary variables that constitute the four main dietary patterns identified

Food items/groups	Principal Component (Dietary Pattern)			
	C 1	C2	C 3	C4
English	0.769	−0.073	0.028	0.212
Sweets sauces	0.738	0.021	0.097	−0.078
Legumes	0.676	0.004	0.456	−0.177
Fast food	0.630	0.103	−0.200	0.337
SSB	0.540	0.027	0.087	0.245
Chocolate	0.506	0.340	−0.150	−0.223
Fried potato	0.501	0.328	0.257	0.356
Other potato	0.391	0.380	0.377	0.246
Sweets and sugar	0.382	0.304	−0.093	0.290
Diet sweet beverages	0.357	0.033	−0.075	−0.091
Dairy	−0.067	0.789	0.181	0.051
Cakes/Pastry/Buns	0.296	0.707	0.323	0.109
Fats and oils	−0.013	0.643	0.071	0.018
Other grains	0.235	0.600	0.145	0.453
Eggs	−0.231	0.592	0.022	−0.119
Biscuits	0.341	0.556	−0.161	−0.116
High salt snacks	0.364	0.493	0.011	0.160
Vegetables	0.006	0.109	0.780	0.047
Nuts	0.126	0.162	0.634	−0.136
Wholegrains/Wholemeal	−0.083	0.065	0.560	−0.067
Fruit	−0.203	−0.095	0.551	−0.007
Fish	−0.144	0.096	0.495	0.296
Savoury sauces	0.152	0.156	0.492	0.153
Pizza	0.192	−0.126	0.337	−0.142
Kebab	−0.005	−0.022	−0.110	0.654
Chinese	0.237	0.121	0.105	0.654
Red Meat	0.057	0.056	−0.044	0.537
Indian	0.172	0.163	0.403	0.537
Poultry	−0.136	−0.113	−0.016	0.366

NB: Major loadings for each item are in red (factor loadings > 0.3 or < −0.3 are considered significant). Extraction method: Principal component analysis. Rotation method: Varimax.



NB: Blue line = the 'Western' dietary pattern (DP1); orange line = the 'Snacks' pattern (DP2); grey line = 'Healthier' pattern (DP3); yellow line = the 'Takeaway and Meat' pattern (DP4)

Figure 6-8 Radar graph of dietary patterns and factor loadings derived through PCA on 29 FFQ food groups (n = 151)

6.4.3.2 24 h 'Multiple Pass' Dietary Recall (24 h MPR)

PCA on 33 food groups was used to analyse habitual and takeaway food intake of 151 participants from 24 h MPR data. The suitability of PCA was assessed prior to analysis. Inspection of the correlation matrix showed that most variables had at least one correlation coefficient greater than 0.25, those that did not have a correlation of >0.25 were removed from the analysis (other potato, Indian, kebab, English takeaway, savoury sauces, fish, alcohol and tea), leaving a remainder of 25 food groups to re-test. The overall Kaiser-Meyer-Olkin (KMO) measure was 0.63 with individual KMO measures > 0.6 and < 0.7, classified as 'mediocre' according to Kaiser (Kaiser, 1974). Bartlett's Test of Sphericity (Bartlett, 1954) reached statistical significance significant ($p < 0.0005$), supporting the factorability of the correlation matrix.

PCA revealed nine components that had eigenvalues > 1, which explained 14.5%, 9.2%, 7.5%, 7.2%, 6.9%, 5.3%, 5.1%, 4.4% and 4.1% of the total variance, respectively. Nevertheless, visual inspection of the scree plot (Appendix 9.38) indicated to retain five components (Cattell, 1966). In addition, a five-component solution met the interpretability criterion. The five-component solution explained 45.3% of the total variance.

To aid the interpretation of these five components an orthogonal (varimax) rotation was performed (Table 6.6). The first component, accounting for 14.5% of the total variance had high positive (> 0.3) factor loadings for dairy (0.725), buns/cakes/pastries (0.682), refined grains (0.672), pizza takeaway (0.540), cocoa (0.407), coffee (0.388), sugar including honey (0.315), oils and dairy free spreads (0.334) and SSB or cordial (0.378). Therefore, this pattern could be described as a ‘Western’ dietary pattern. There were no negative loadings > −0.3.

The second component in the rotated structure matrix, accounting for 9.2%, of the total variance, could be identified as a ‘Cosmopolitan’ dietary pattern with high positive (> 0.3) loadings obtained for egg (0.621), wholegrains/meal (0.593), fruit (0.542), coffee (0.520), vegetables (0.451) and high negative (> −0.3) loadings for high-salt snacks (−0.476), sugar (−0.404) and SSB or cordial (−0.304) (Table 6.6).

The third component, accounting for 7.5% of the total variance, was characterised by high positive (> 0.3) loadings for fast food (0.685), potato fried (0.671), poultry (0.509), oils and dairy free spreads (0.476), SSB or cordial (0.456) and Chinese takeaway (0.384) (Table 6.6). There were no negative loadings > −0.3. Thus, this pattern could be described as ‘Takeaway Food'.

The fourth component, accounting for 7.2% of the total variance, was identified as a ‘Prudent’ pattern and presented high positive (> 0.3) loadings for legumes (including soy) (0.792), nuts and seeds (0.632), fruit juice (0.567), vegetables (0.565), and high negative (> −0.3) loadings for processed red meat (−0.386) (Table 6.6).

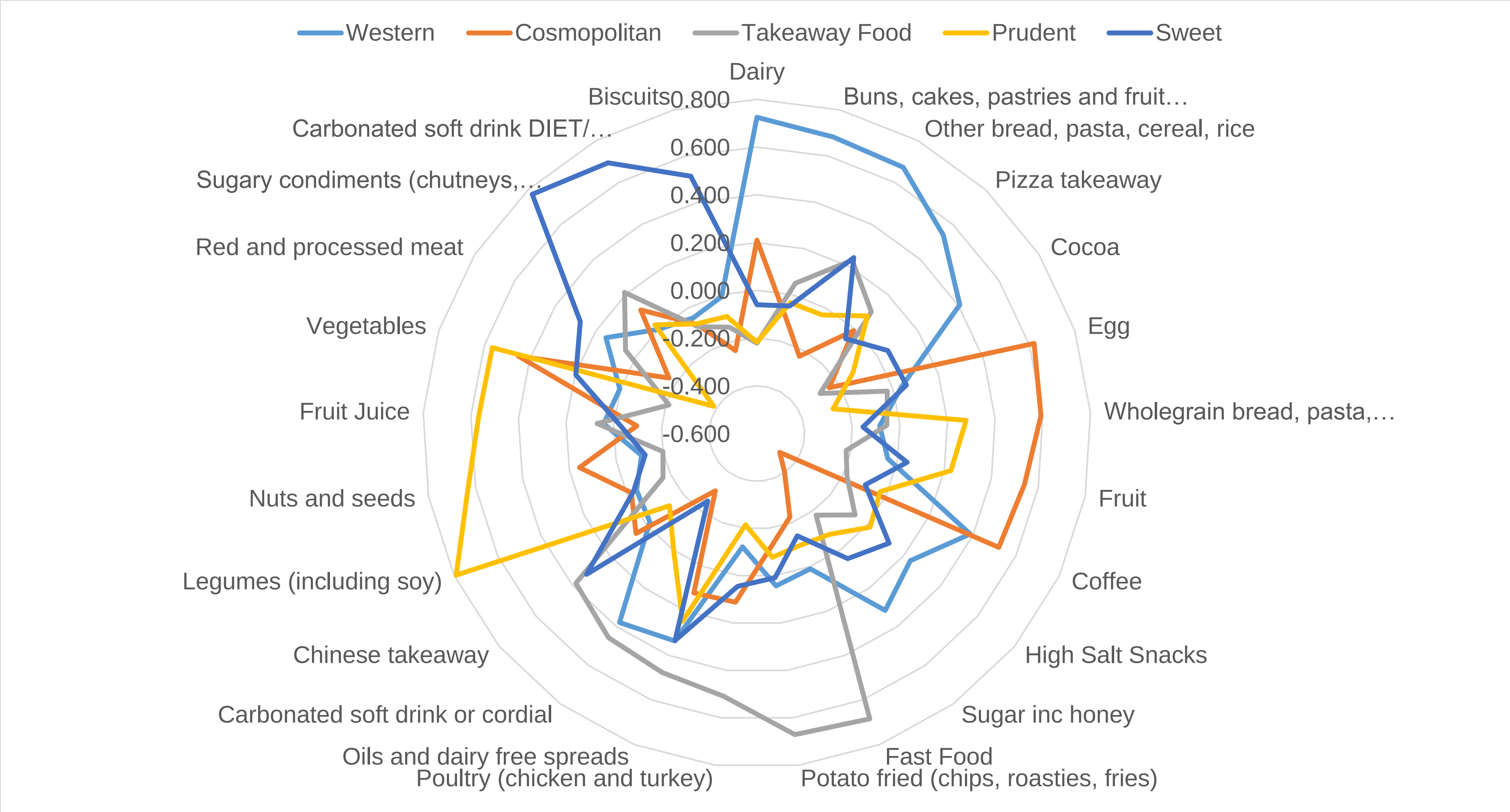
The final fifth pattern to emerge, accounting for 6.9% of the total variance, was a ‘Sweet’ dietary pattern with high positive (> 0.3) factor loadings for sugary condiments (0.774), diet drinks/sweeteners (0.694), biscuits (0.513), oils and dairy free spreads (0.332) and Chinese takeaway (0.325) with no negative loadings > −0.3 (Table 6.6).

Figure 6.9 presents a visual interpretation of the differences between the five dietary patterns produced from the 24 h MPR through PCA.

Table 6-6 Rotated component loadings derived from PCA with Varimax Rotation on 25 food groups from 24 h MPR for dietary variables that constitute the five main dietary patterns identified

Food items/groups	Principal Component (Dietary Pattern)				
	C 1	C 2	C 3	C4	C5
Dairy	0.725	0.211	−0.222	−0.217	−0.060
Buns/Cakes/Pastries	0.682	−0.104	0.051	−0.034	−0.049
Refined grains	0.672	−0.231	0.229	−0.032	0.242
Pizza takeaway	0.540	−0.008	0.100	0.076	−0.056
Cocoa	0.407	−0.241	−0.287	−0.123	0.050
Egg	0.037	0.621	−0.025	−0.264	0.059
Wholegrains/Wholemeal	−0.084	0.593	−0.054	0.278	−0.155
Fruit	−0.041	0.542	−0.219	0.227	0.041
Coffee	0.388	0.520	−0.182	−0.029	−0.097
High-salt snacks	0.235	−0.476	−0.066	0.015	0.120
Sugar inc honey	0.315	−0.404	−0.177	−0.079	0.048
Fast food	0.010	−0.224	0.685	−0.098	−0.139
Potato fried	0.043	−0.122	0.671	−0.078	0.009
Poultry	−0.122	0.112	0.509	−0.215	0.045
Oils/Dairy free spreads	0.334	0.118	0.476	0.245	0.332
SSB or cordial	0.378	−0.304	0.456	−0.006	−0.250
Chinese takeaway	−0.019	0.057	0.384	−0.125	0.325
Legumes (inc soy)	−0.043	−0.019	−0.165	0.792	−0.028
Nuts and seeds	−0.108	0.156	−0.199	0.632	−0.124
Fruit juice	0.043	−0.096	0.071	0.567	−0.025
Vegetables	0.003	0.451	−0.214	0.565	0.197
Processed red meat	0.149	−0.165	0.051	−0.386	0.276
Sugary condiments	0.000	0.110	0.210	0.025	0.774
Diet drinks/Sweeteners	−0.047	−0.076	−0.089	−0.075	0.694
Biscuits	−0.008	−0.241	−0.140	−0.094	0.513

NB: Major loadings for each item are in red (factor loadings > 0.3 or < −0.3 are considered significant). Extraction method: Principal component analysis. Rotation method: Varimax.



NB: Blue line = the 'Western' dietary pattern (DP1); orange line = the 'Cosmopolitan' pattern (DP2); grey line = the 'Takeaway Food' pattern (DP3); yellow line = the 'Prudent' pattern (DP4); dark blue line = the 'Sweet' pattern (DP5).

Figure 6-9 Radar graph of dietary patterns and factor loadings derived through PCA on 25 24 h MPR food groups (n = 151

6.4.4 METABOLITE FINGERPRINTS OF TAKEAWAY FOOD CONSUMERS

6.4.4.1 FOODS/FOOD GROUPS OF SPECIFIC INTEREST

PCA (unsupervised method) reduced data dimensionality and PC-LDA (supervised method) followed. PC-LDA was performed using both positive and negative ionisation modes of data to produce different ionisation adducts and signals, this was to enable the best representation of different metabolite fingerprints. The PC-LDA Eigenvalue (Tw), defined as the ratio of the between and within-group standard deviations on the discriminant variables, was used to evaluate the performance of PC-LDA. The larger the Tw value, the greater the discriminatory power of the model. Discrimination was considered poor for Tw values < 1, weak for Tw values between 1 and 2, and good for Tw values > 2 (Enot et al., 2008).

The results showed that there was good separation of metabolite patterns from participants reporting a high (> 4.0) consumption of specific takeaway foods in three days, others reporting a low (< 2.0) consumption, and non-consumers. Specific takeaway foods with a good classification included fast food (in positive and negative modes), Chinese (positive mode), Indian (positive and negative modes), English (positive and negative modes), and kebab (positive and negative modes) (Table 6.7). Food groups with a poor classification were total takeaway food, pizza and red and processed meat (Table 6.7). Plots of the first two discriminant functions (DFs), DF1 and DF2, of each analysis allowed visualisation of the goodness-of-class separation. There were no results for discriminant function 3 (DF3) due to < 3 groups in the class structure.

Table 6-7 PC-LDA Eigenvalues (Tw) for takeaway food categories

	Fast Food	Chinese takeaway	Indian takeaway	English takeaway	Pizza takeaway	Kebab takeaway	TAKE-AWAY	Red and processed
posDF1	3.3	3.0	3.7	4.6	1.4	7.2	1.3	0.5
posDF2	1.2	2.4	2.7	1.4	0.9	2.9	0.9	0.4
posDF3							0.4	0.2
negDF1	3.2	1.8	6.1	7.1	1.0	7.0	1.8	0.8
negDF2	2.7	1.1	2.8	3.8	0.7	2.1	1.2	0.7
negDF3							0.6	0.3

NB: Discrimination considered adequate for Tw values > 2.0 are in red. DF = discriminant function. Pos = positive mode. Neg = negative mode.

PCA was used to examine data clustering without any pre-definition of sample class membership. The PCA technique (unsupervised) in the negative mode showed a natural clustering for Indian takeaway food with high discrimination between high consumption and low or non-consumption (Fig. 6.10). PC1 on the x-axis showed the biggest variance (27.1%) of separation for low and non-consumption from high consumption, whereas PC2 on the y-axis showed separation between low and non-consumption with less explained variance (14.2 %) (Fig. 6.10). There were three clear clusters between non/low/high exposure ranges to Indian takeaway food, meaning that there were chemical differences between the metabolite fingerprints. Plots for PCA positive and PCA-LDA positive available for Indian takeaway in Appendix 9.41.

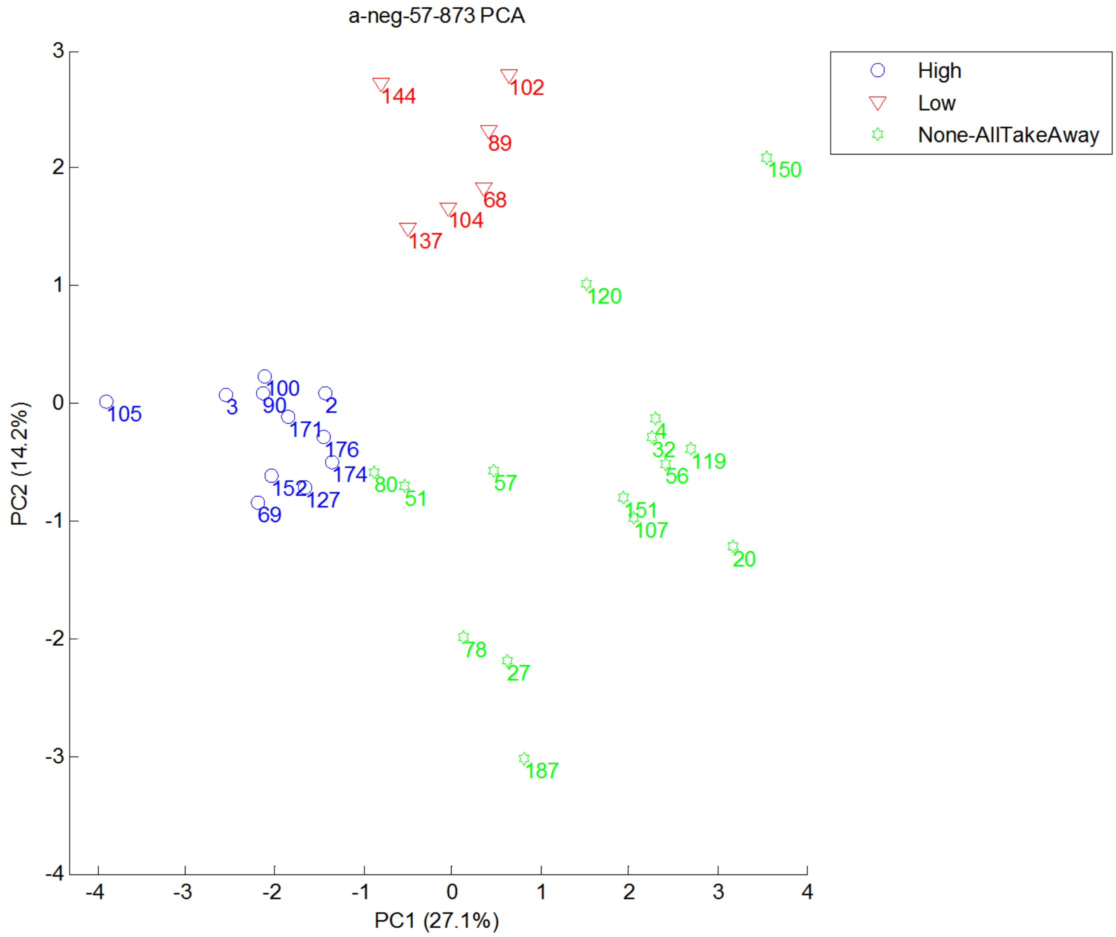


Figure 6-10 PCA Indian takeaway negative mode

PC-LDA (a powerful supervised multivariate analysis) used sample class label information (in this instance non/low/high exposure to Indian) to find the linear combination of variables to best separate the three classes of metabolite fingerprints. The PC-LDA negative mode showed most variance between Tw values for Indian takeaway food. Plots of the first two Discriminant Functions (DFs) were used to visualize the goodness of class separation and DF1 separated all three classes (non/low/high). The scree plot shows that on DF1 on the x-axis, with an eigenvalue (Tw) value of 6.09 in the dimension of maximal discrimination, high Indian consumption was adequately discriminated from low and non-consumption (Fig. 6.11). A Tw value > 2 provided confidence there were chemical differences between the metabolite fingerprints of non/low/high classes for Indian and that further investigation was potentially worthwhile.

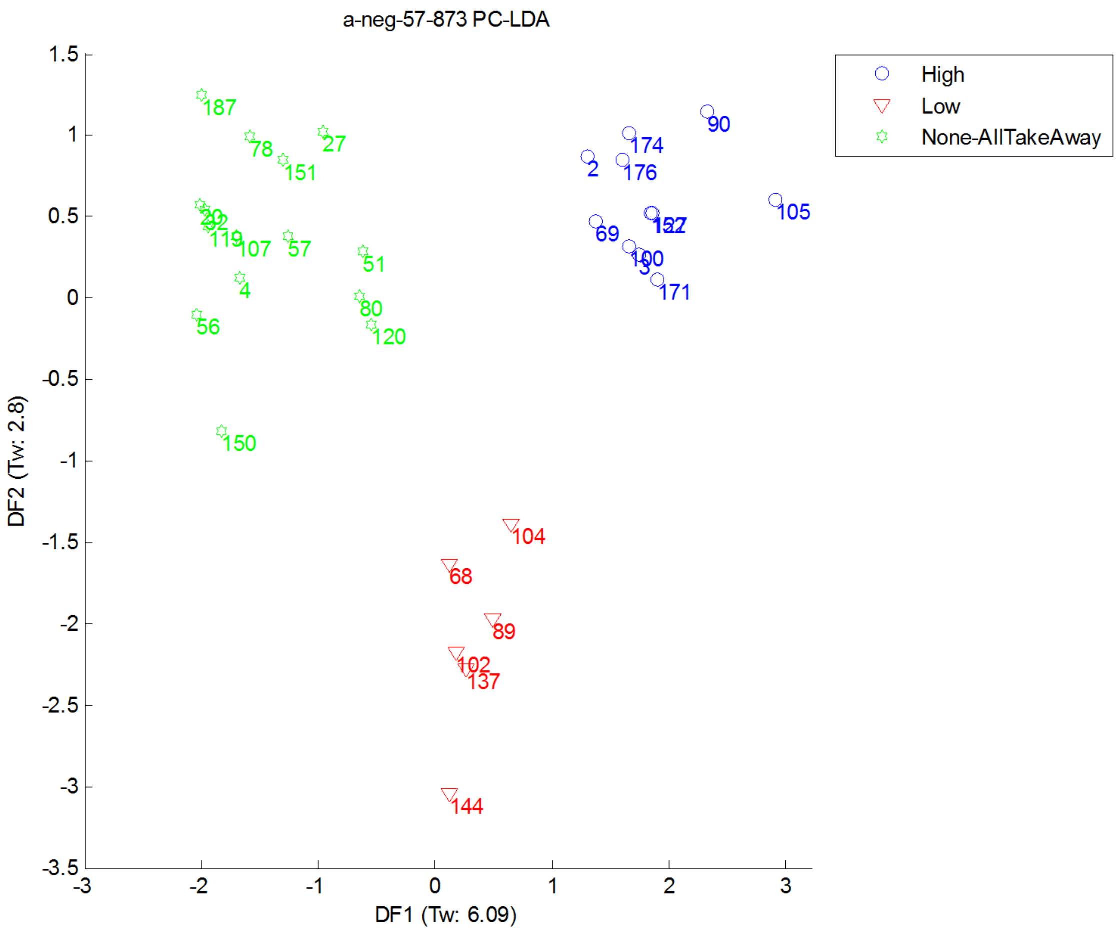


Figure 6-11 PC-LDA plot Negative mode m/z 57-873 for Indian takeaway food

PC-LDA (supervised analysis) used sample class label information (non/low/high exposure to fast food) to find the linear combination of variables to best separate the three classes of metabolite fingerprints. PC-LDA in the negative mode showed most variance between Tw values for fast food. With a Tw value of 2.66 on DF2 (Fig. 6.12) it is evident that high consumption of fast food were adequately discriminated from low and non-consumption of fast food. A Tw value > 2 provided confidence that there were chemical differences between the metabolite fingerprints of non/low/high classes for fast food and that further investigation was potentially worthwhile. Plots for PCA negative, PCA positive and PCA-LDA positive available for fast food in Appendix 9.42.

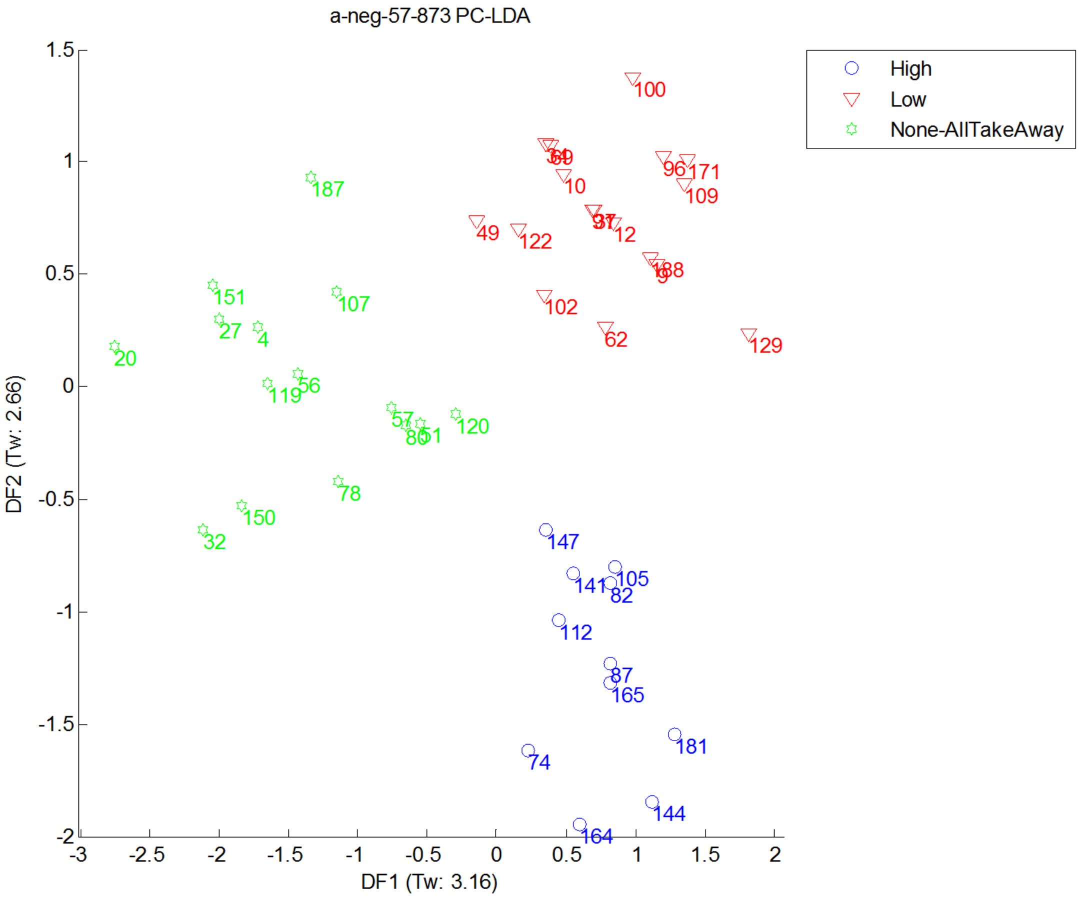


Figure 6-12 PC-LDA plot Negative mode m/z 57-873 for fast food

PC-LDA (supervised analysis) used sample class label information (non/low/high exposure to Chinese takeaway food) to find the linear combination of variables to best separate the three classes of metabolite fingerprints. The positive mode showed most variance between Tw values for Chinese takeaway food. High Chinese takeaway food consumption was adequately discriminated from low and non-consumption of Chinese based on a Tw value of 2.36 on DF2 (Fig. 6.13). A Tw value > 2 provided confidence that there were chemical differences between the metabolite fingerprints of non/low/high classes for Chinese and that further investigation was potentially worthwhile. Plots for PCA negative, PCA-LDA negative and PCA positive available for Chinese takeaway in Appendix 9.43.

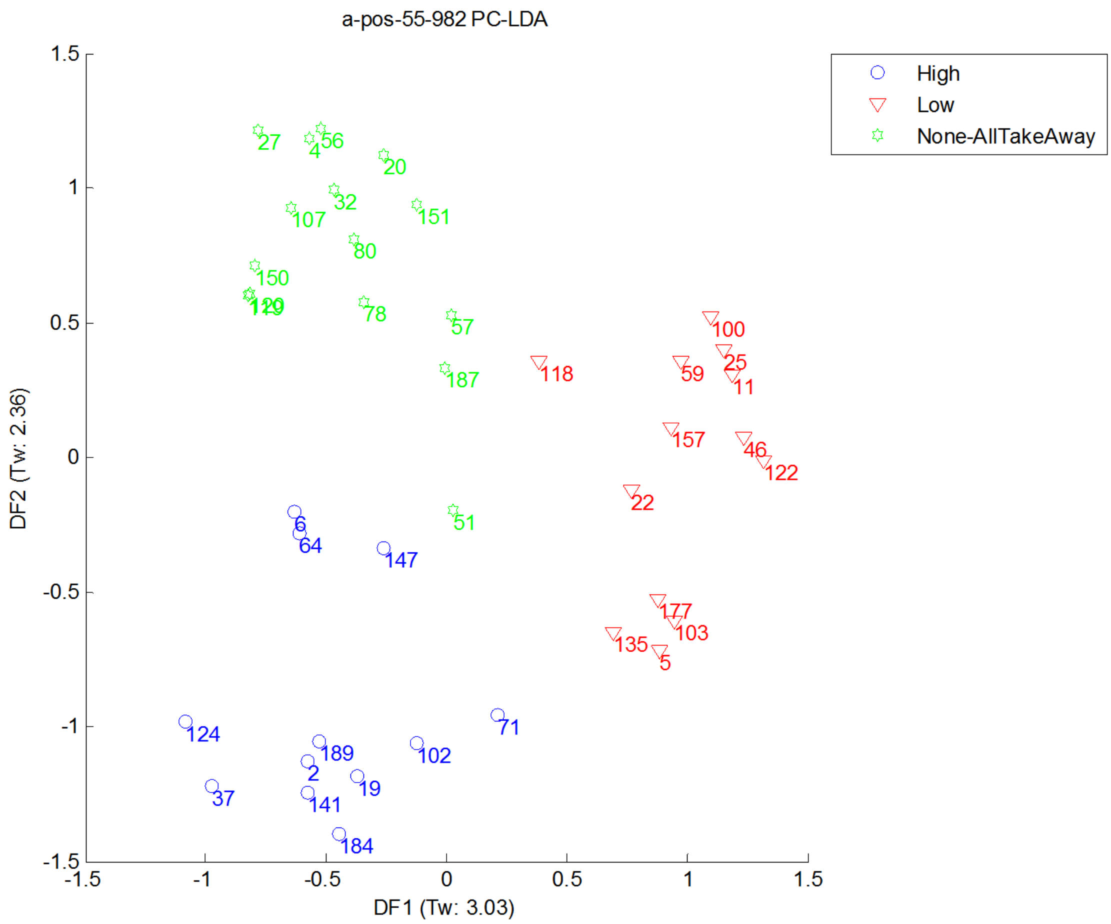


Figure 6-13 PCA-LDA plot Positive mode m/z 55 – 982 for Chinese takeaway

PC-LDA (supervised analysis) used sample class label information (non/low/high exposure to English takeaway) to find the linear combination of variables to best separate the three classes of metabolite fingerprints. The negative mode showed most variance between Tw values for English takeaway food. With a Tw value of

3.76 on DF2 (Fig. 6.14) the scree plot shows that high consumption of English takeaway food were adequately discriminated from low and non-consumption. A Tw value > 2 provided confidence that there were chemical differences between the metabolite fingerprints of non/low/high classes for English takeaway food and that further investigation was potentially worthwhile. Plots for PCA negative, PCA positive and PCA-LDA positive available for English takeaway in Appendix 9.44.

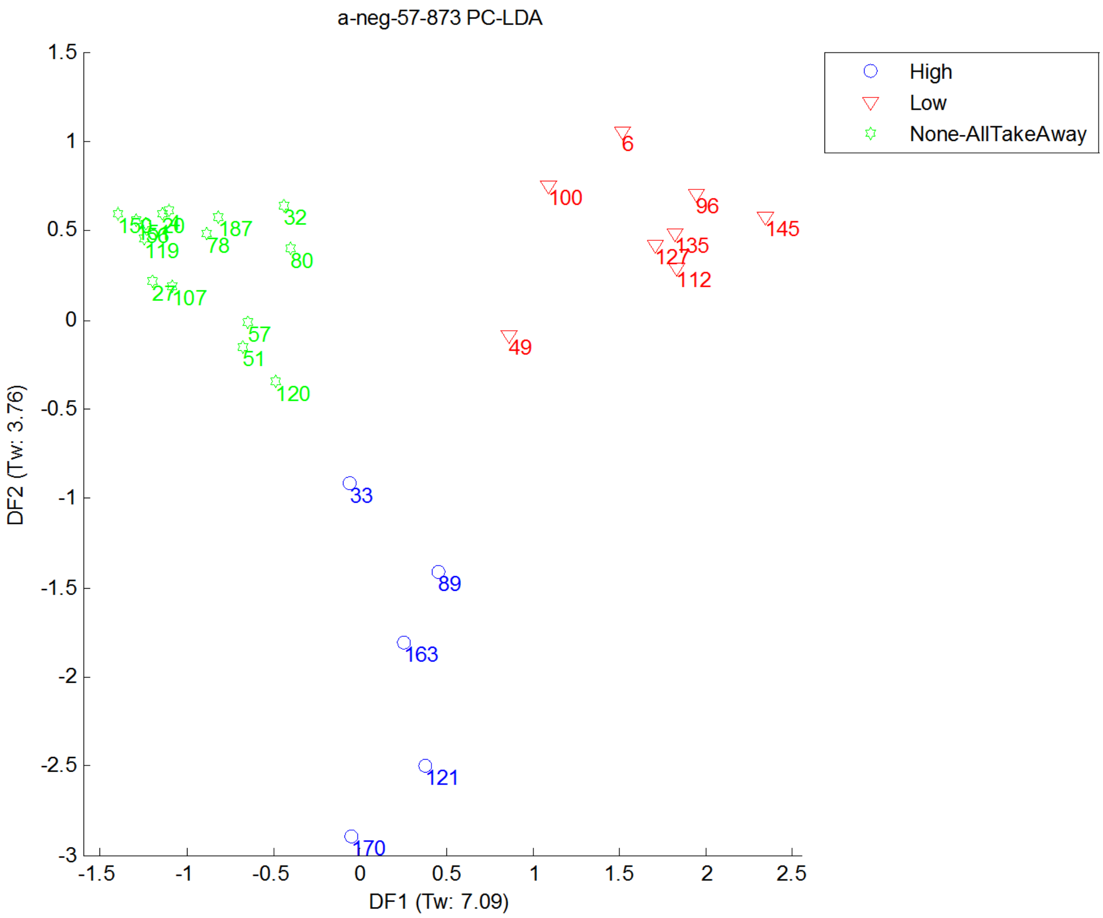


Figure 6-14 PC-LDA plot Negative mode m/z 57-873 for English takeaway

PC-LDA (supervised analysis) used sample class label information (non/low/high exposure to kebab) to find the linear combination of variables to best separate the three classes of metabolite fingerprints. The negative mode showed most variance between Tw values for kebab. High kebab consumption was adequately discriminated from low and non-consumption of kebabs based on a Tw value of 6.96 (Fig. 6.15). A Tw value > 2 provided confidence that there were chemical differences between the metabolite fingerprints of non/low/high classes for kebab and that further investigation was potentially worthwhile. Plots for PCA negative, PCA positive and PCA-LDA positive available for kebab in Appendix 9.45.

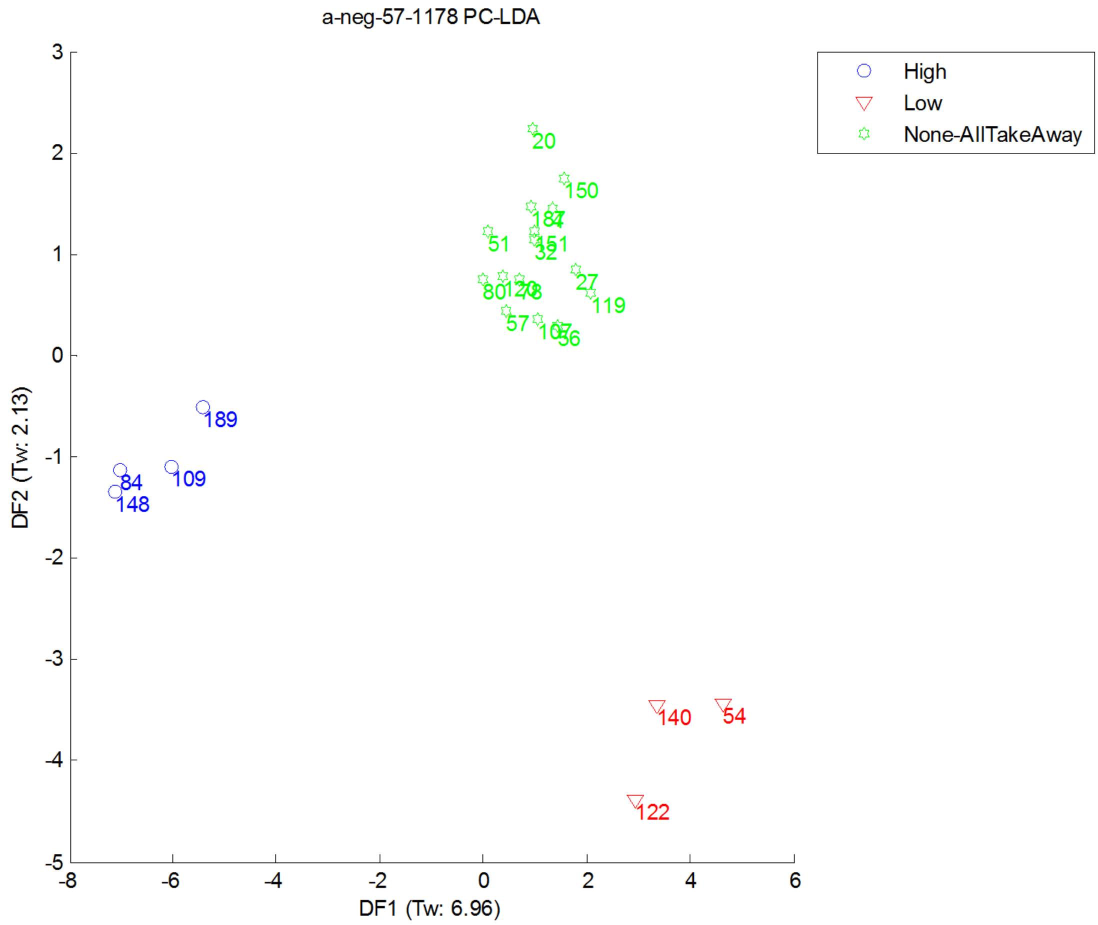


Figure 6-15 PC-LDA plot Negative mode m/z 57-873 for kebab

In addition, plots for PCA negative, PCA positive, PCA-LDA negative and PCA-LDA positive are available for pizza, not included in the results, in Appendix 9.46.

Figure 6.16 shows a scores plot from a PC-LDA of FIE-HRMS fingerprints representing urine samples from participants classified by non/low//high/very-high exposure ranges for all (total) takeaway food. The negative mode showed most variance between Tw values for total takeaway food. There were three clear clusters with good discrimination shown for very high consumers and non-consumers and a mixed cluster for low and high consumption. The mixed cluster suggests that the metabolite fingerprints of all the samples were similar and thus were difficult to discriminate. Despite this blurred cluster, there is a crude gradation between low and high, moving from negative to positive on DF1 (Fig. 6.16). However, the Tw value of 1.77 in the dimension of maximal discrimination suggests that very high and high total takeaway food were poorly discriminated from low and non-consumption of total takeaway food. Plots for PCA negative, PCA positive and PCA-LDA positive available (Appendix 9.47).

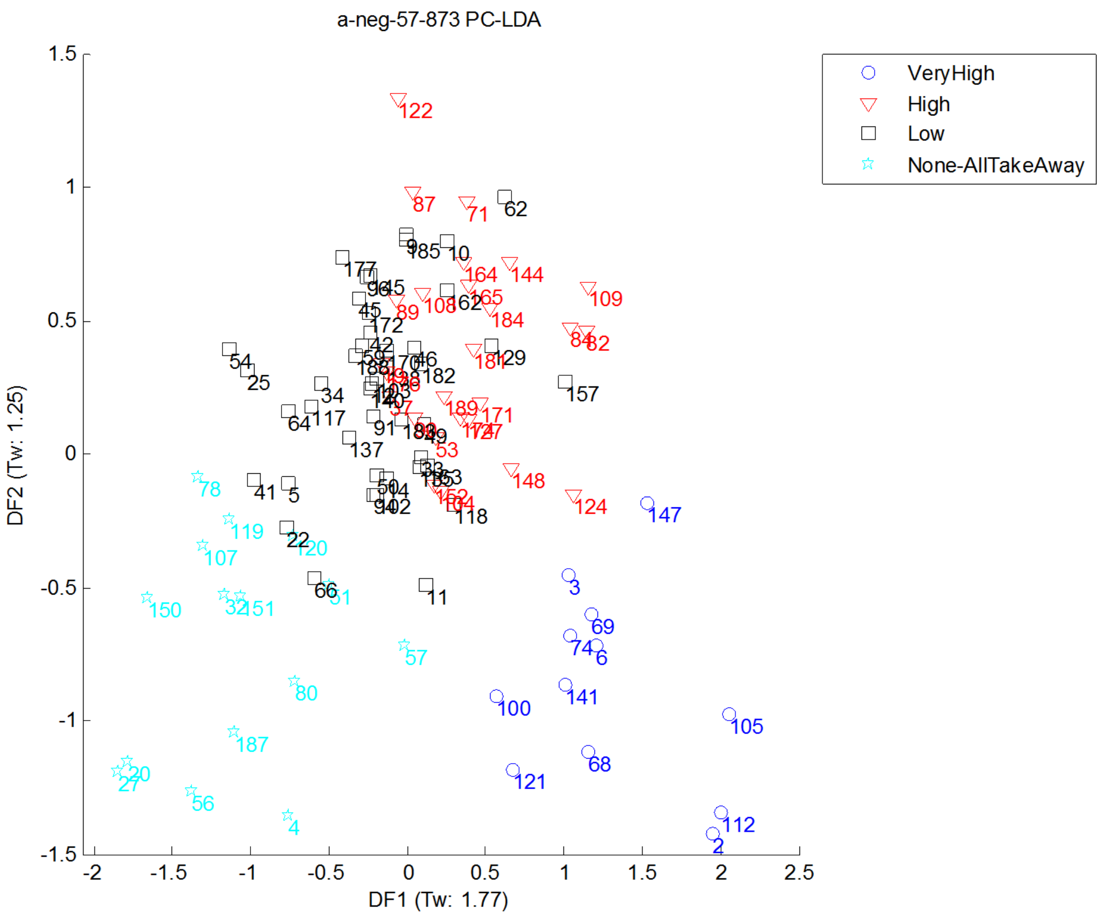


Figure 6-16 PC-LDA plot DF1 vs DF2 Negative mode m/z 57-873 for all takeaway food

6.4.4.2 TARGET FOODS FROM 24 H MPR DIETARY PATTERNS

Poultry (chicken or turkey) consumption was a class structure by which PC-LDA indicated differences between high and non-consumption, while low consumption showed overlap with non- and high consumption (Fig. 6.17). The Tw value was 1.44 indicating a weak discrimination (< 2.0 Tw threshold) for poultry however, there was a trend in separation on DF1 (Fig 6.17). The signals that were influencing this split and causing discrimination were then identified. One of the top ranked signals causing the discrimination between high and non-consumption of poultry was 3- Methylhistidine (Fig. 6.17), appearing three times, as two adducts and one isotope (Table 6.8).

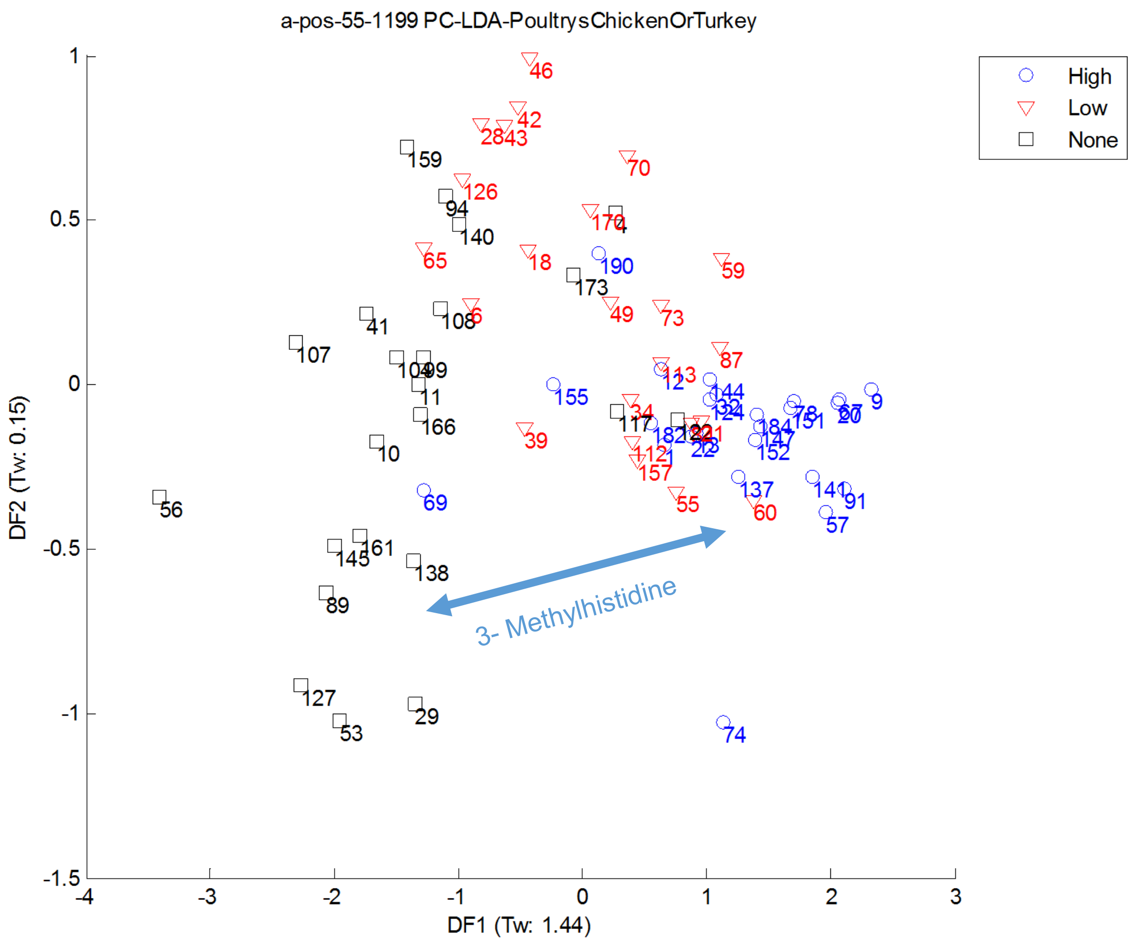


Figure 6-17 Signals for habitual dietary poultry (chicken or turkey)

In Table 6.8 *m/z* signals were linked to a specific metabolite for poultry by atomic mass information. The positive mode showed that participants consuming high quantities of poultry had high levels of 3-Methylhistidine represented by two ionisation products; M+H and M+Na adducts and a 13C isotope. Therefore, 3-Methylhistidine was the top ranked signal correlated with poultry exposure.

Table 6-8 Identity of highly ranked and correlated signals explanatory of poultry exposure

Food Item	ANOVA-F	p-value	*m/z*	class 1	class 2	Mode	Ionisation product	Accurate mass	ID
Poultry	29.980	2.40381E-06	170.091942	High	None	Pos	$[M+H]^{1+}$	170.092404	3-Methylhistidine
Poultry	18.124	0.000117486	171.0951659	High	None	Pos	$[M+H]^{1+}$ 13C	171.095758	3-Methylhistidine
Poultry	17.051	0.000174284	192.0738626	High	None	Pos	$[M+Na]^{1+}$	192.074348	3-Methylhistidine

NB: Pos = positive.

Coffee consumption was a class structure by which PC-LDA indicated differences between high, low and non-consumption (Fig. 6.18). The Tw value was 2.71 and showed that poultry was adequately discriminated (> 2.0 Tw threshold) with a good separation on DF1 (Fig 6.18). One of the top ranked signals influencing this split and causing the discrimination between high, low and non-consumption of coffee in the negative mode was Quinic acid ($[M-H]^{1-}$, *m/z* 191.0561134) (Fig. 6.18; Table 6.9). Whereas, Trigonelline (N-methyl nicotinate) (*m/z* 138.05479, $[M+H]^{1+}$ and 13C isotope) were highly ranked in positive mode (Table 6.9).

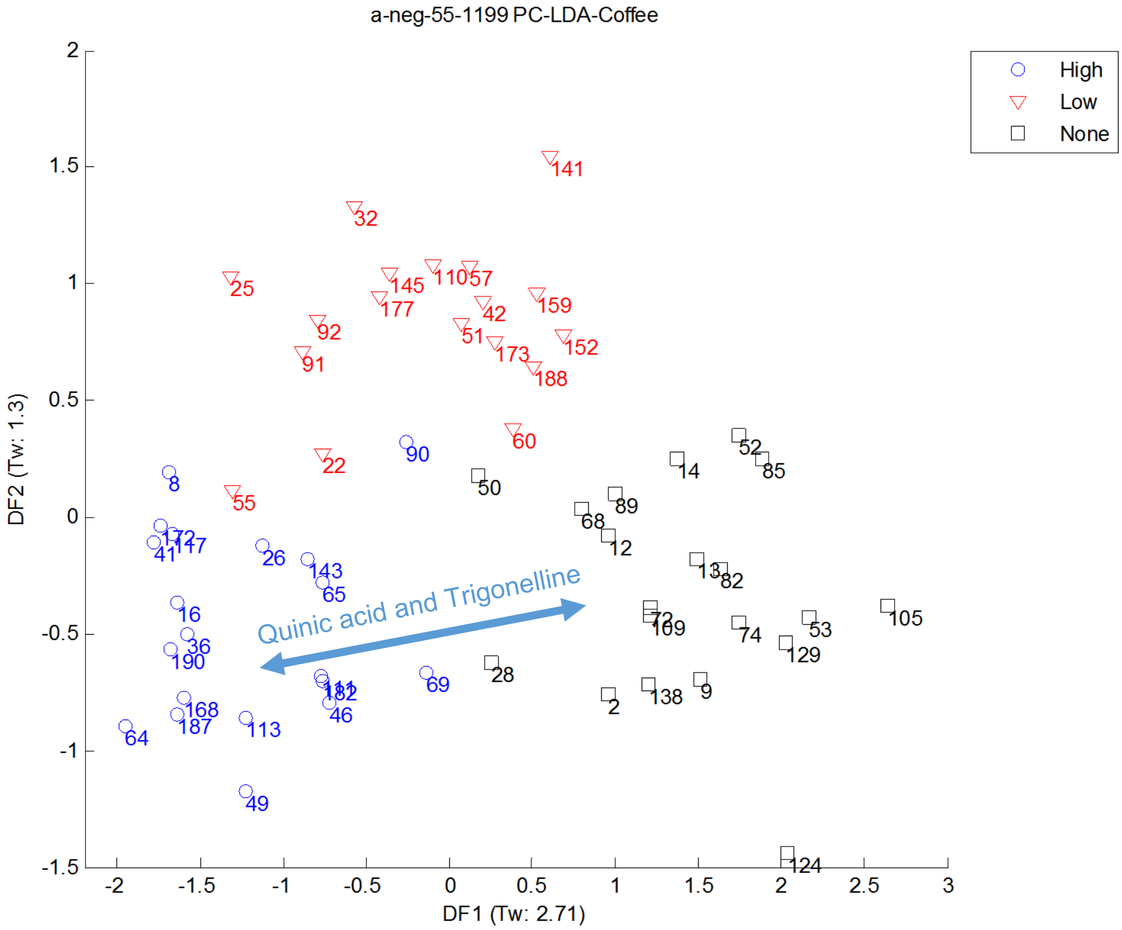


Figure 6-18 Signals for habitual dietary coffee

In Table 6.9 *m/z* signals were linked to specific metabolites for coffee by atomic mass information. The positive mode showed that participants consuming high quantities of coffee had high levels of Trigonelline. Whereas the negative mode showed high consumption of coffee as Quinic acid. Trigonelline and Quinic acid were the top ranked signals correlated with coffee exposure.

Table 6-9 Identity of highly ranked and correlated signals explanatory of coffee exposure

Food Item	ANOVA-F	p-value	*m/z*	class 1	class 2	Mode	Ionisation product	Accurate mass	ID
Coffee	79.846	8.98278E-11	191.0561479	High	None	Neg	$[M-H]^{1-}$	191.0561134	Quinic acid
Coffee	23.187	2.63844E-05	138.0546413	High	None	Pos	$[M+H]^{1+}$	138.05479	Trigonelline (N-methyl nicotinate)

Coffee	21.810	4.09766E-05	139.0579531	High	None	Pos	13C [M+H]$^{1+}$	139.05823	Trigonelline (N-methyl nicotinate)13C
Coffee	19.873	8.14667E-05	191.0561479	Low	None	Neg	[M-H]$^{1-}$	191.0561134	Quinic acid

NB: Pos = positive. Neg = negative.

Alcohol consumption was a class structure by which PC-LDA indicated differences between high and low or non-consumption (Fig. 6.19). The Tw value was 4.32 and showed that alcohol was adequately discriminated (> 2.0 Tw threshold) with a good separation on DF1 (Fig 6.19). A top ranked signal causing the discrimination between high and low or non-consumption of alcohol (wine and beer combined), beer and wine was Ethyl glucuronide (Fig. 6.19).

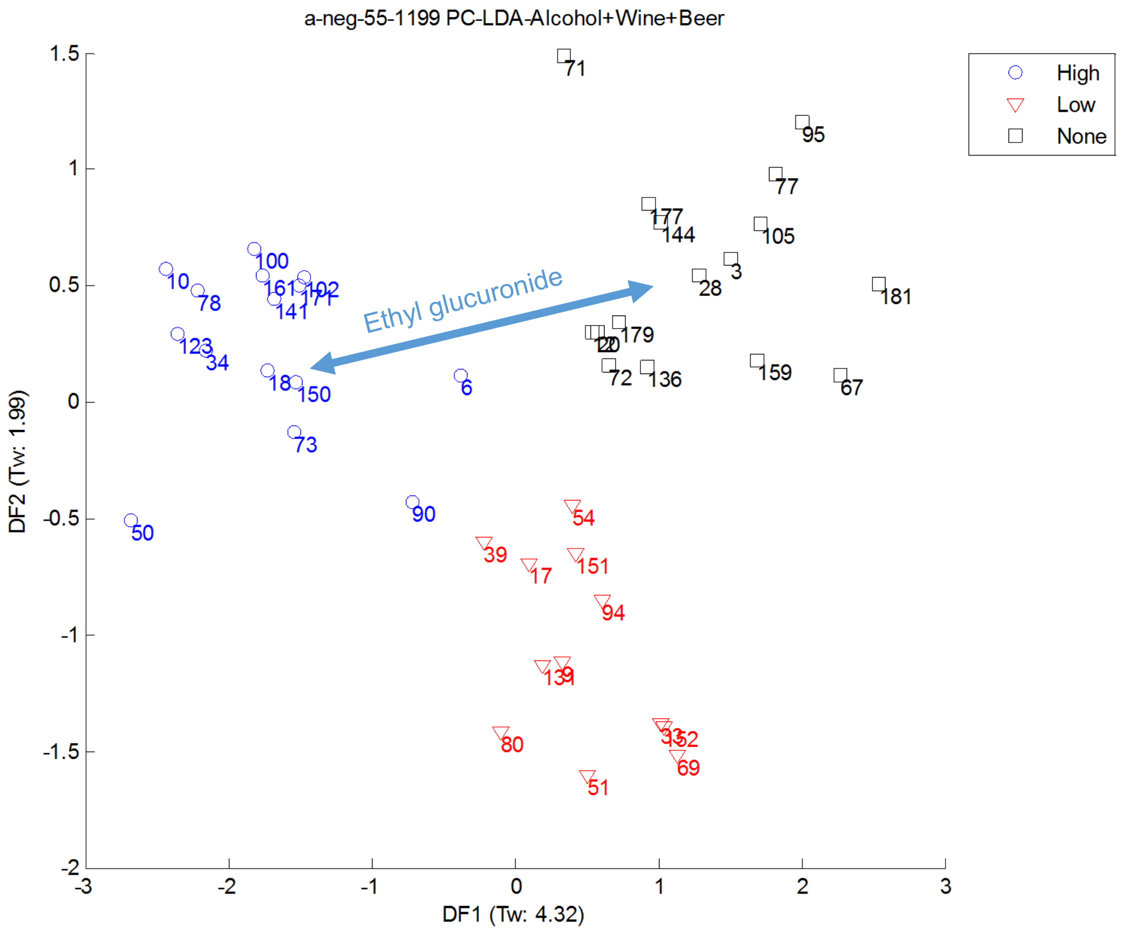


Figure 6-19 Signals for habitual dietary alcohol

In Table 6.10 *m/z* signals were linked to a specific metabolite for alcohol by atomic mass information. The negative mode showed that participants consuming high quantities of alcohol/wine/beer had high levels of Ethyl glucuronide and 2-Isopropylmalic acid, the top ranked signals correlated with alcohol exposure.

Table 6-10 Identity of highly ranked and correlated signals explanatory of alcohol exposure

Food Item	ANOVA-F	p-val	*m/z*	class 1	class 2	Mode	Ionisation product	Accurate mass	ID
Alcohol/Wine/Beer	89.587	3.19364E-10	221.0668414	High	None	Neg	$[M-H]^{1-}$	221.0666784	Ethyl glucuronide
Alcohol/Wine/Beer	36.936	2.3744E-06	221.0668414	High	Low	Neg	$[M-H]^{1-}$	221.0666784	Ethyl glucuronide
Alcohol/Wine/Beer	30.206	7.16112E-06	175.0610402	High	None	Neg	$[M-H]^{1-}$	175.0611984	2-Isopropylmalic acid
Beer	42.064	1.43543E-05	221.0668414	High	None	Neg	$[M-H]^{1-}$	221.0666784	Ethyl glucuronide
Beer	21.399	0.000392606	175.0610402	High	None	Neg	$[M-H]^{1-}$	175.0611984	2-Isopropylmalic acid
Wine	151.247	3.08832E-09	175.0610402	High	None	Neg	[M-H]1-	175.0611984	2-Isopropylmalic acid
Wine	24.285	9.33975E-05	175.0610402	Low	None	Neg	[M-H]1-	175.0611984	2-Isopropylmalic acid
Wine	31.015	0.000121995	221.0668414	High	Low	Neg	[M-H]1-	221.0666784	Ethyl glucuronide
Wine	21.285	0.000337811	221.0668414	High	None	Neg	[M-H]1-	221.0666784	Ethyl glucuronide

NB: Neg = negative.

Wine consumption (as opposed to alcohol) was a class structure by which PC-LDA indicated differences between high and low or non-consumption (Fig. 6.20). The Tw value was 9.56 and showed that wine was adequately discriminated (> 2.0 Tw threshold) with a good separation on DF1 (Fig 6.20). A top ranked signal causing the discrimination between high and low or non-consumption of wine, but not general alcohol or beer, was Tartaric acid (Fig. 6.20).

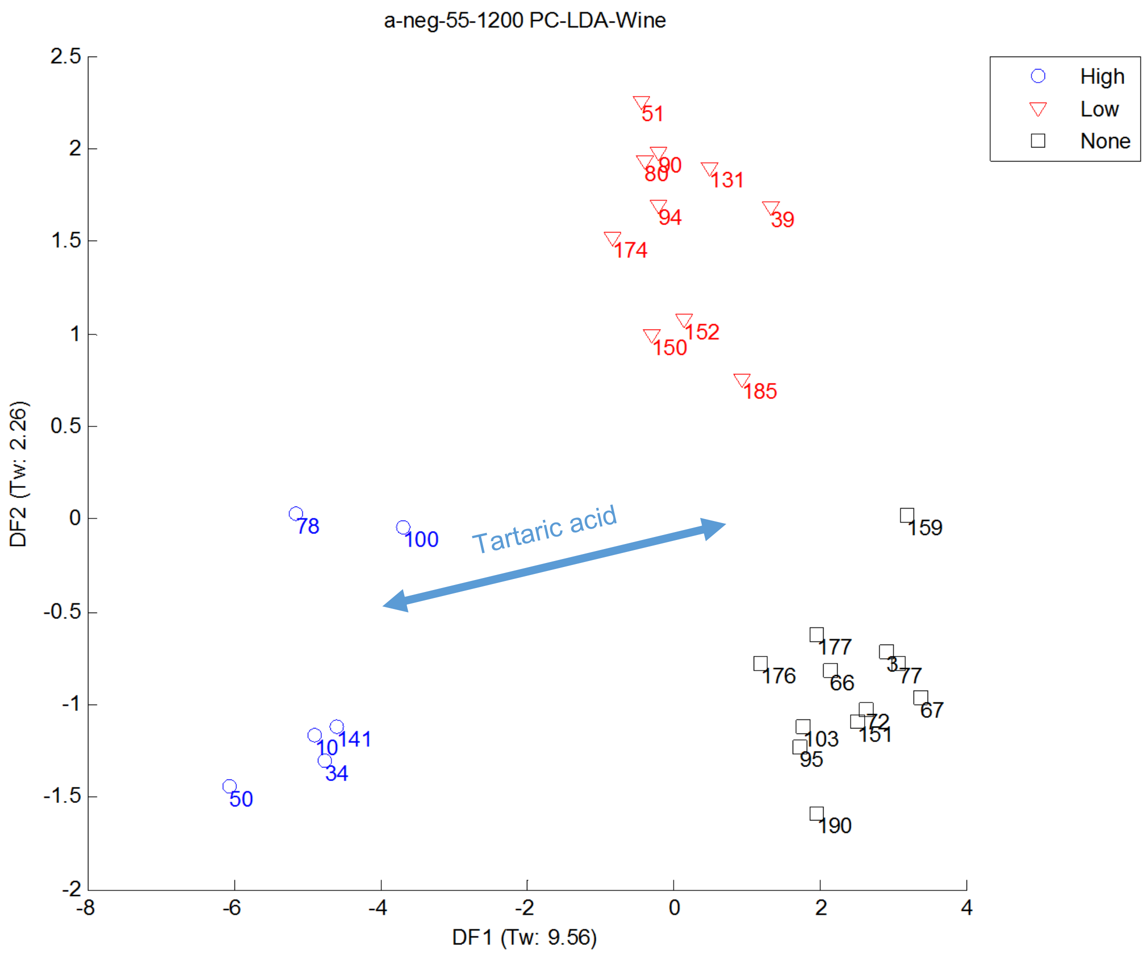


Figure 6-20 Signals for habitual dietary wine

In Table 6.11 *m/z* signals were linked to a specific metabolite for wine by atomic mass information. The negative mode showed that participants consuming high quantities of wine had high levels of Tartaric acid, the top ranked signal correlated with wine exposure.

Table 6-11 Identity of highly ranked and correlated signals explanatory of wine exposure

Food Item	ANOVA-F	p-value	*m/z*	class 1	class 2	Mode	Ionisation product	Accurate mass	ID
Wine	15.578	0.001938515	149.0088923	High	Low	Neg	$[M-H]^{1-}$	149.00916	Tartaric acid
Wine	24.920	0.000160845	149.0088923	High	None	Neg	$[M-H]^{1-}$	149.00916	Tartaric acid
Wine	15.784	0.00184977	150.0122468	High	Low	Neg	$[M-H]^{1-}$13C	150.01252	Tartaric acid

Wine	23.232	0.000224926	150.0122468	High	None	Neg	$[M-H]^{1-}$ 13C	150.01252	Tartaric acid

NB: Neg = negative.

6.4.5 METABOLITE FINGERPRINTS OF HABITUAL DIETARY PATTERN GROUPS

6.4.5.1 FINGERPRINTS FROM 24 H MPR DATA

Urine samples from participants scoring high on each of the five dietary patterns from 24 h MPR were analysed to determine if metabolite fingerprints could classify the dietary patterns. The fingerprint clusters in Figure 6.21 represent groups of individuals scoring highest on the five dietary patterns in the un-rotated structures produced from PCA on the 24 h MPR (Appendix 9.39). There was a slight visible gradient from cluster 1 (red) in the negative to cluster 2 (lime green) and cluster 3 (emerald green) in the positive mode (Fig. 6.21). Despite very low Tw values < 1 (Table 6.12) from PC-LDA, which suggests poor discrimination due to much overlap of the dietary patterns (Fig. 6.21), there did appear to be discrimination between classes 1 and 2 (dietary patterns 1 and 2) and between 1 and 3 (dietary patterns 1 and 3).

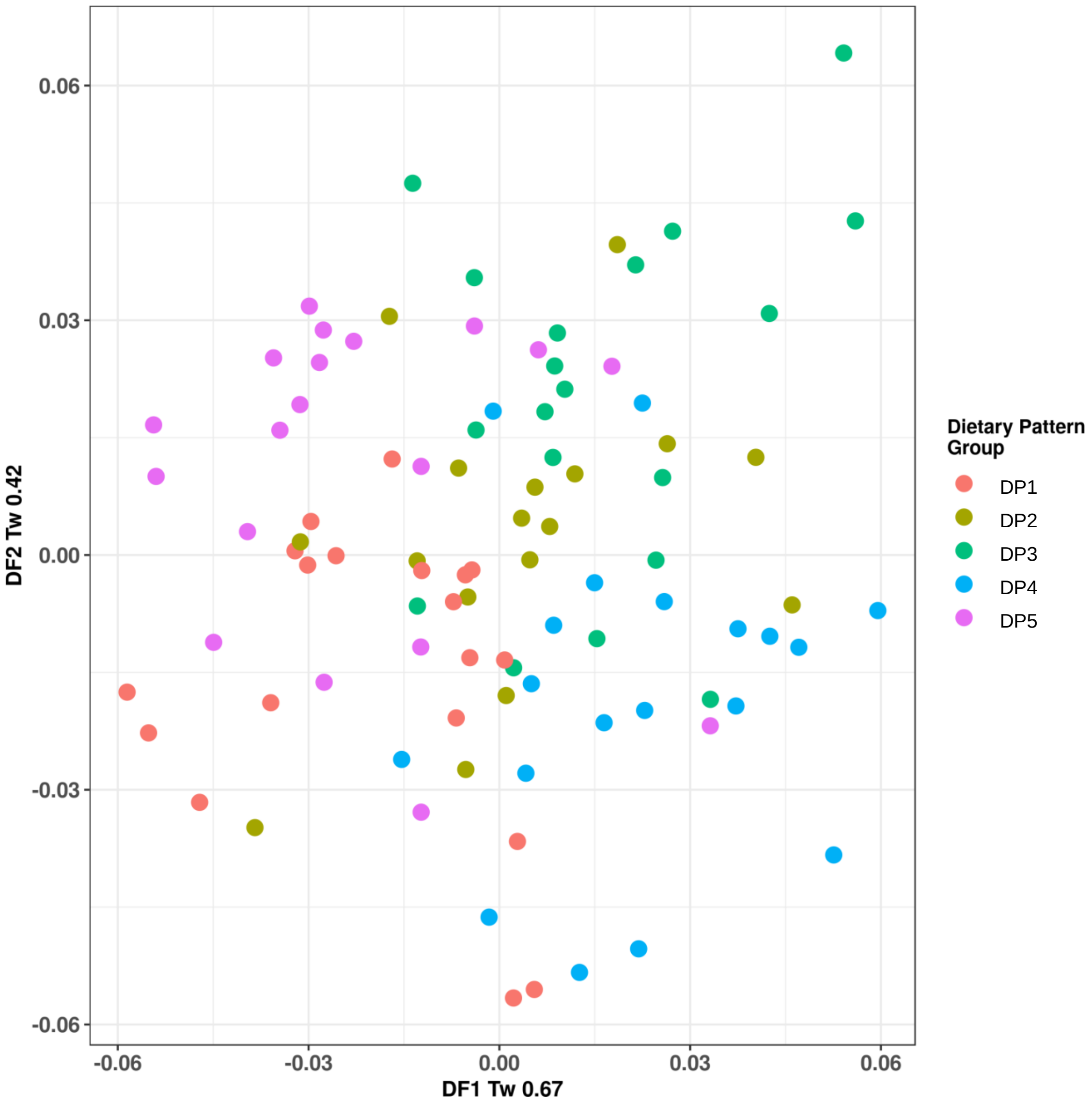


Figure 6-21 Metabolite fingerprints from 3 x 24 h MPR

Table 6-12 PC-LDA Eigenvalues (Tw) for dietary patterns from 3 x 24 h MPR

	Eig (Tw)	Perceig
DF1	0.665	50.083
DF2	0.416	31.341
DF3	0.148	11.181
DF4	0.098	7.396

Accuracy, margins of classification and area under the ROC (Receiver Operator Characteristic) curve (AUC) were used to evaluate the robustness and performance of classification models. Models can be deemed adequate overall if random forest (RF) margins > 0.2 and AUC values > 0.8 (Lloyd et al., 2013). Pairwise comparisons were made for all five classes/dietary patterns (e.g. DP1 versus DP2 etc.). The discrimination between classes 1 and 2 (DP1 and DP2) and between 1 and 3 (DP1 and DP3) was indicated by supervised RF margin valves > 0.2 (Table 6.13). These pairwise comparisons between the two sets of classes were investigated further to identify the top ranked signals causing the discrimination.

Table 6-13 Performance of classification models from 3 x 24 h MPR

Pairwise	Margin	Accuracy	AUC
1~2	0.22	0.75	0.84
1~3	0.28	0.78	0.91
1~4	0.10	0.67	0.75
1~5	0.08	0.65	0.66
2~3	0.03	0.56	0.64
2~4	0.08	0.66	0.73
2~5	-0.03	0.42	0.40
3~4	0.12	0.65	0.74
3~5	0.05	0.58	0.65
4~5	0.15	0.69	0.79

Ultra High Performance Liquid Chromatography-High Resolution MS (UHPLC-HRMS) and Tandem mass spectrometry (MSn) identified the structures of putative biomarkers. The top ranked signals causing the discrimination between classes 1 and 2 (DP1 and DP2) was 3 or 7-Methylxanthine (Table 6.14). This is a biomarker of cocoa consumption. When comparing dietary pattern data from the 24 h MPR to urine samples, cocoa consumption from chocolate scored highest in dietary pattern 2 in the PCA un-rotated component matrix (Table 6.15) and likewise was highest in dietary pattern 2 for relative intensity in urine samples (Fig. 6.22) therefore validating results.

Table 6-14 Identity of signal potentially explanatory of cocoa exposure

	Pairwise	Feature	Score	P value	Putative ID	Putative MF, inoisation product and accurate mass
HR24 DP	1~2	167.0558	12.19	4.09E-05	3 or 7-Methylxanthine	$C_6H_6N_4O_2$ & $[M+H]^{1+}$ = 167.0563526

Table 6-15 PCA un-rotated component matrix for cocoa

Dietary pattern	1	2	3	4	5
Cocoa	0.253	0.419	−0.050	−0.254	0.133

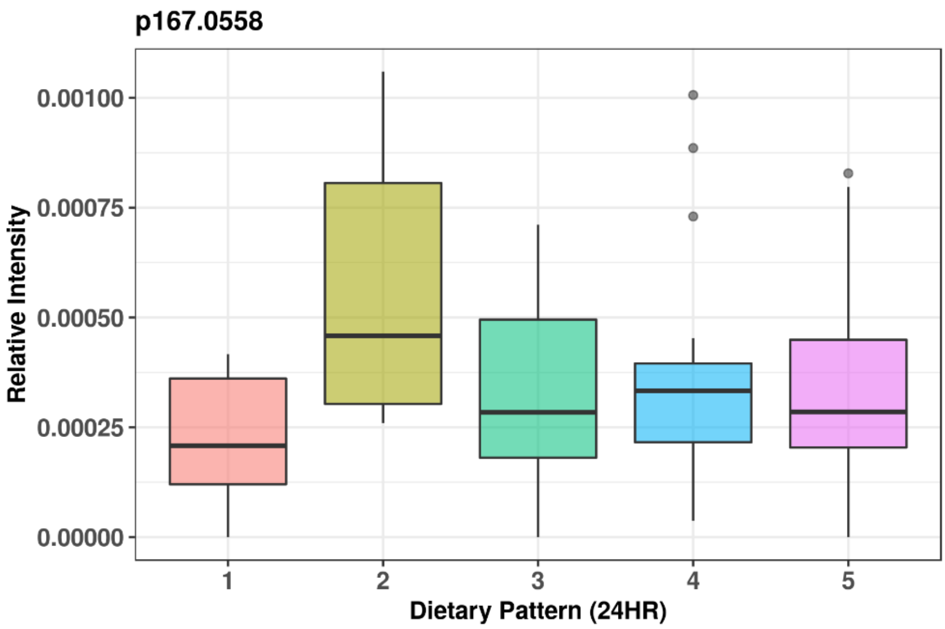


Figure 6-22 3 or 7-Methylxanthine semi quantitative concentration

A top ranked signal causing the discrimination between classes 1 and 3 (DP1 and DP3) was Trigonelline (N-methyl nicotinate) (Table 6.16). This is a known biomarker of coffee consumption. When comparing dietary patterns from the 24 h MPR to urine samples, coffee scored highest in dietary pattern 2 in the PCA un-rotated component matrix (Table 6.17) and in dietary pattern 3 from relative intensity (Fig. 6.23). However, lowest scores for coffee correlated between 24 h MPR and urine samples in dietary patterns 1 and 5, presenting a degree of validity.

Table 6-16 Identity of signal potentially explanatory of coffee exposure

	Pair wise	Feature	Score	P value	Putative ID	Putative MF, inoisation product and accurate mass
HR24 DP	1~3	160.03638	9.32	0.0025991	Trigonelline (N-methyl nicotinate)	$C_7H_7NO_2$ & $[M+Na]^{1+}$ = 160.0369003
HR24 DP	1~3	138.05466	10.54	0.002452	Trigonelline (N-methyl nicotinate)	$C_7H_7NO_2$ & $[M+H]^{1+}$ = 138.0549556
HR24 DP	1~3	139.05792	9.88	0.0011591	Trigonelline (N-methyl nicotinate)	$C_7H_7NO_2$ & $[M+H]^{1+}$ ^{13}C = 139.05807

Table 6-17 PCA un-rotated component matrix for coffee

Dietary pattern	**1**	**2**	**3**	**4**	**5**
Coffee	−0.210	0.484	0.201	0.023	−0.381

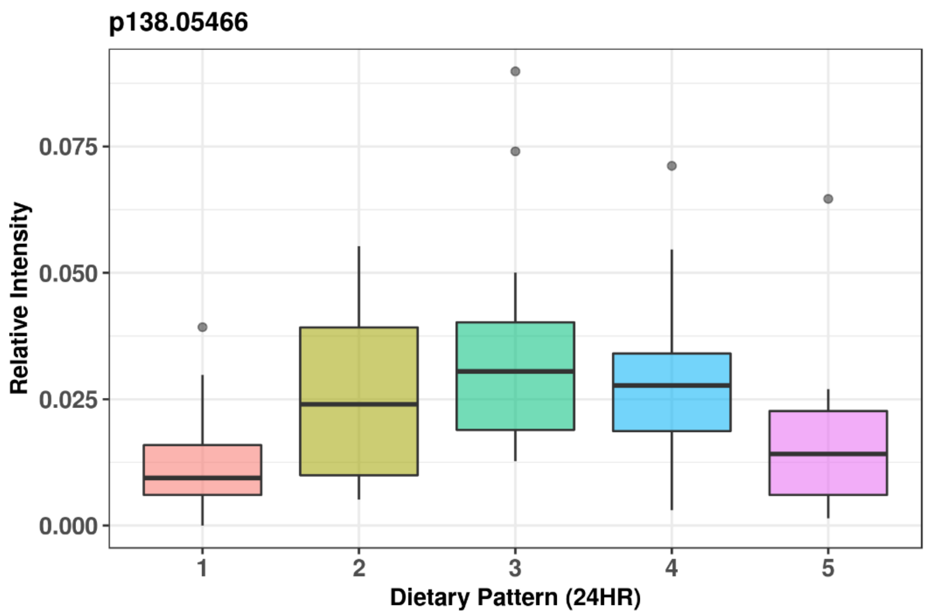


Figure 6-23 Trigonelline semi quantitative concentrations

A top ranked signal causing the discrimination between classes 1 and 3 (DP1 and DP3) was Tyrosol glucuronide (Table 6.18). This is a biomarker for wine and virgin olive oil consumption. When comparing dietary patterns from the 24 h MPR to urine samples, oils and dairy free spreads scored highest in dietary pattern 3 in the PCA un-rotated component matrix (Table 6.19) and for relative intensity in urine samples (Fig. 6.24).

Table 6-18 Identity of signal potentially explanatory of wine and virgin olive oil exposure

	Pairwise	Feature	Score	P value	Putative ID	Putative MF, inoisation product and accurate mass
HR24 DP	1~3	337.09	7.87	0.00744	Tyrosol glucuronide	$C_8H_{10}O_2$ & $[M+Na]^{1+}$ = 161.0573013

Table 6-19 PCA un-rotated component matrix for oils and dairy free spreads

Dietary pattern	1	2	3	4	5
Oils and dairy free spreads	0.237	0.121	0.346	0.194	−0.034

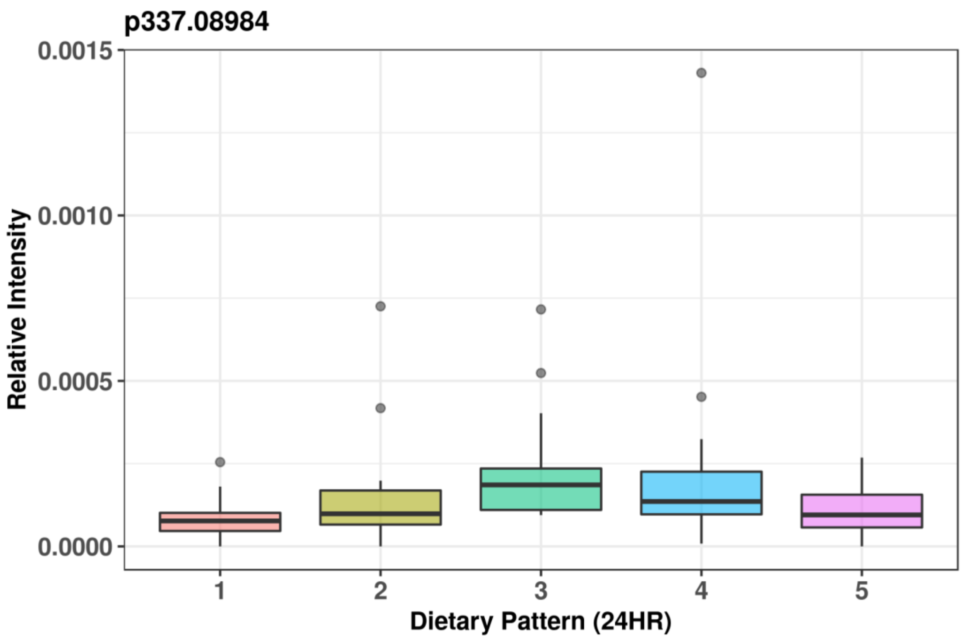


Figure 6-24 Tyrosol glucuronide.

Another top ranked signal causing the discrimination between classes 1 and 3 (DP1 and DP3) was Hippuric acid (Table 6.20). This is a general biomarker of total vegetable and fruit juice consumption. When comparing dietary patterns from the 24 h MPR to urine samples, vegetables scored highest in dietary pattern 3 in the PCA un-rotated component matrix (Table 6.21) and for relative intensity in urine samples (Fig. 6.25). Likewise, fruit scored highest in dietary pattern 3 from both dietary measures (24 h MPR and urine).

Table 6-20 Identity of signal potentially explanatory of fruit, vegetable and fruit juice exposure

	Pairwise	Feature	Score	P value	Putative ID	Putative MF, inoisation product and accurate mass
HR24 DP	1~3	178.05081	11.43	4.04E-04	Hippuric acid	$C_9H_9NO_3$ & $[M-H]^{1-}$ = 178.0509674
HR24 DP	1~3	179.05423	14.21	7.63E-05	Hippuric acid	$C_9H_9NO_3$ & $[M-H]^{1-}$ ^{13}C = 179.0543214

Table 6-21 PCA un-rotated component matrix for vegetables and fruit

Dietary pattern	**1**	**2**	**3**	**4**	**5**
Vegetables	−0.597	0.247	0.288	0.271	0.184
Fruit	−0.530	0.169	0.256	0.071	−0.128

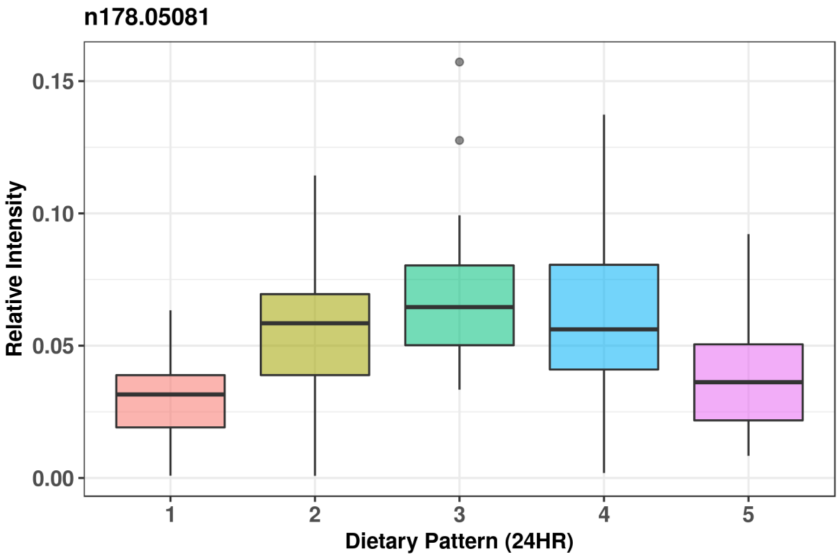


Figure 6-25 Hippuric acid

6.4.5.2 Fingerprints from FFQ data

Urine samples from participants scoring high on each of the four dietary patterns from the FFQ were analysed to determine if metabolite fingerprints could be used to classify these dietary patterns. The fingerprint clusters in Figure 6.26 represent groups of individuals scoring highest on the four dietary patterns in the un-rotated structures produced from PCA on the FFQ (Appendix 9.40). There is a slight visible gradient across from cluster 1 (red) in the negative to cluster 2 (green) in the positive, which separates DP1 (a Western dietary pattern) to DP2 (a Prudent dietary pattern) (Fig. 6.26). As with the 24 h MPR data, even with very low Tw values from PC-LDA (Table 6.22) there appeared to be some discrimination between classes 1 and 2 with RF margin valves ≥ 2.0 (Table 6.23) showing discrimination between the two dietary patterns. This pairwise comparison was investigated further to identify the top ranked signals causing the discrimination.

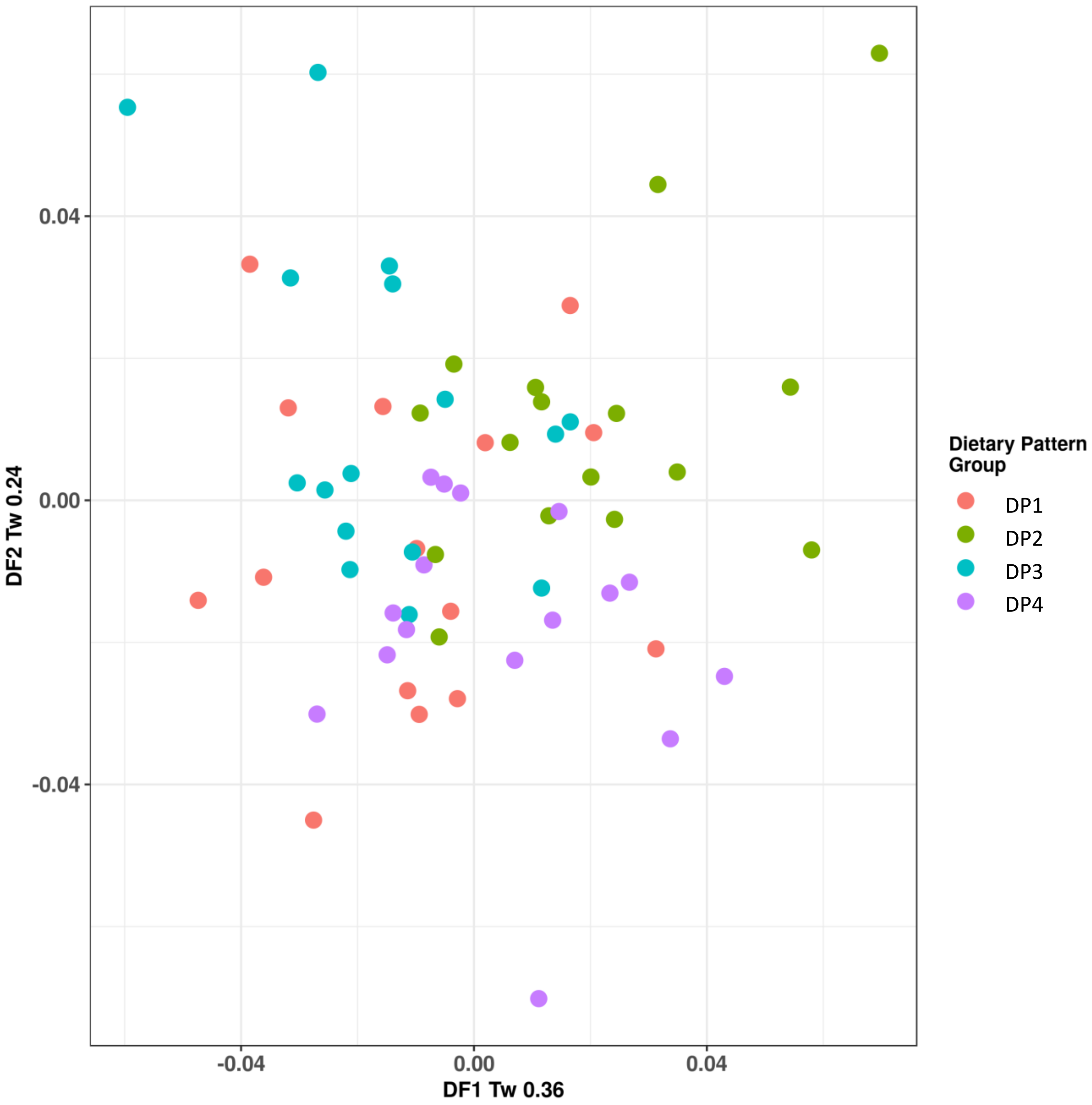


Figure 6-26 Metabolite fingerprints from FFQ

Table 6-22 PC-LDA Eigenvalues (Tw) for dietary patterns from FFQ

	Eig (Tw)	Perceig
DF1	0.365	49.428
DF2	0.239	32.337
DF3	0.135	18.235

Accuracy, margins of classification and area under the ROC (Receiver Operator Characteristic) curve (AUC) were used to evaluate the robustness and performance of classification models. Models were deemed adequate overall if random forest (RF) margins > 0.2 and AUC values > 0.8 (Lloyd et al., 2013). Pairwise comparisons were made for all four classes/dietary patterns (e.g. DP1 versus DP2). The pairwise comparison between classes 1 and 2 were investigated further to identify the top ranked signals causing the discrimination (Table 6.23).

Table 6-23 Performance of classification models from FFQ

Pairwise	Margin	Accuracy	AUC
1~2	0.20	0.77	0.86
1~3	0.01	0.55	0.58
1~4	0.09	0.64	0.69
2~3	0.03	0.56	0.63
2~4	0.01	0.52	0.57
3~4	0.01	0.51	0.55

After further analysis, results showed very few putative identities were made for the top ranked signals causing the discrimination between dietary patterns 1 and 2. Potential unhealthy markers (signals A, B and C) were thought to be the cause for the discrimination, in line with previous work (Lloyd, personal communication) (Fig. 6.27; 6.28; 6.29). All three signals displayed different characteristics however, classification of these markers requires further investigation.

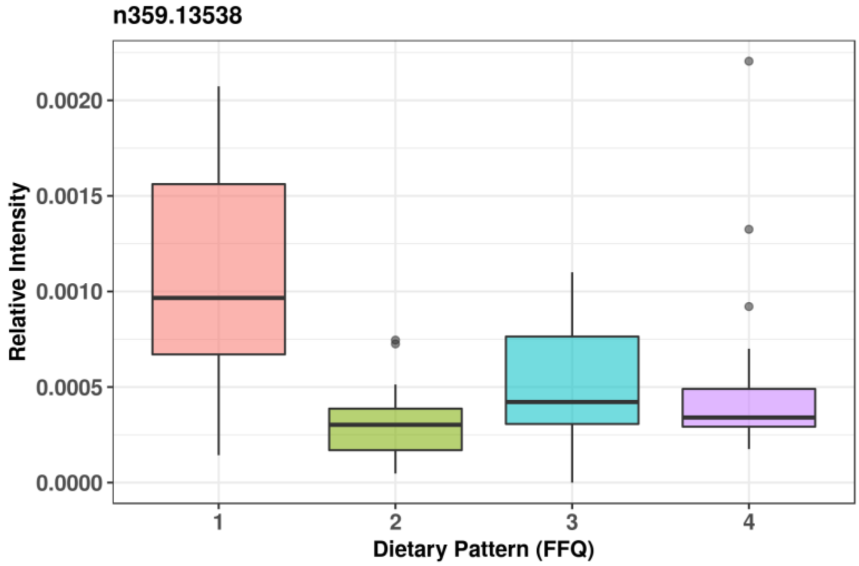


Figure 6-27 Potential unhealthy marker (Signal A)

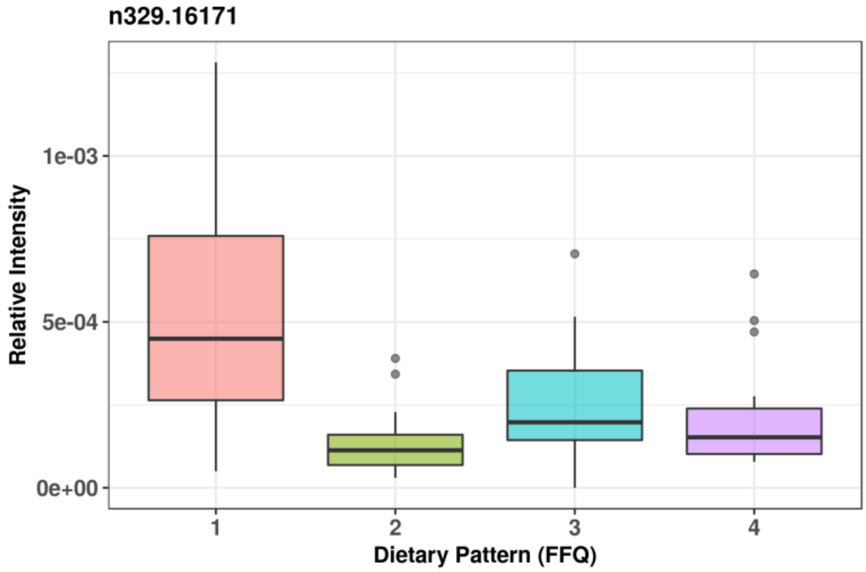


Figure 6-28 Potential unhealthy marker (Signal B)

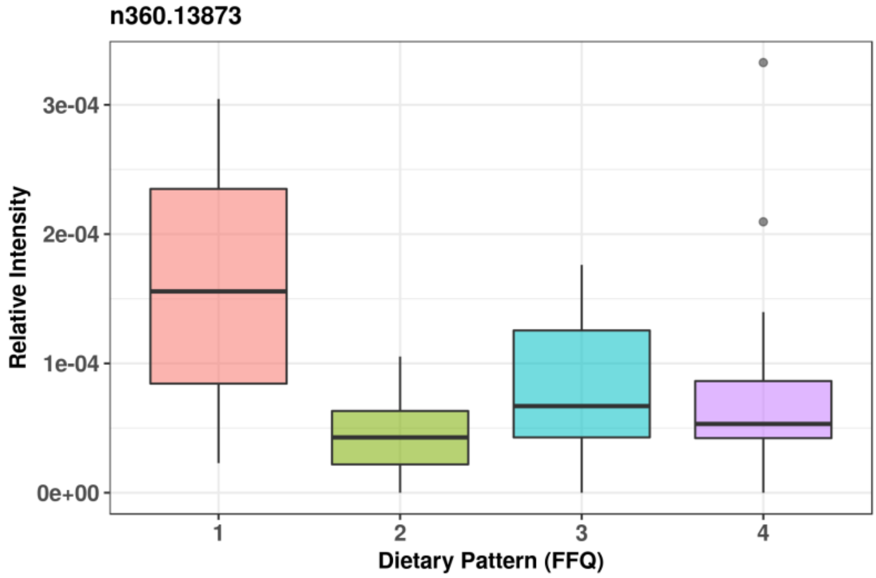


Figure 6-29 Potential unhealthy marker (Signal C)

6.5 Discussion

6.5.1 Validity of a food frequency questionnaire for identifying dietary patterns among self-reported takeaway food consumers

In the present study, the validity of the EHS as a dietary assessment tool was tested by producing dietary patterns from the data, which were compared to those from 3 x 24 h MPR. PCA produced four patterns from the EHS, these were classified as; 1) Western, 2) Snacks, 3) Healthier and 4) Takeaways and Meat (Table 6.5), and five dietary patterns from the 24 h MPR, classified as; 1) Western, 2) Cosmopolitan 3) Takeaway 4) Prudent and 5) Sweet (Table 6.6). Figure 6.8 and 6.9 showed a degree of similarity between the dietary patterns produced using the two forms of dietary assessment. Both sets of PCA produced a western dietary pattern and a prudent style dietary pattern; however, the food groups included in each of the dietary patterns varied. From the 24 h MPR data

(Table 6.6) the Prudent pattern had high positive (> 0.3) loadings for legumes (including soy), nuts and seeds, fruit juice, vegetables, and high negative (> -0.3) loadings for processed red meat. Whereas in the FFQ data (Table 6.5) the Healthier pattern had high positive (> 0.3) loadings for vegetables, nuts, wholegrains/meal, fruit, fish, savoury sauces, legumes, Indian takeaway, other potato, pizza takeaway, cakes/pastry/buns and no high negative loadings (> -0.3). Results showed that some of the food groups in the dietary patterns overlap in types of foods consumed from both dietary assessment techniques; the 24 h MPR and FFQ; supporting the argument that the 24 h MPR validated dietary patterns from the EHS (FFQ). For the first dietary pattern (defined as the Western pattern) in both analysis there was a crossover of chocolate, potato and sugar (Fig. 6.8; 6.9). For dietary pattern two (defined as Cosmopolitan from the 24 h MPR and Snacks from the FFQ) egg scored highly. These findings validate the FFQ and would indicate that there is a slight degree of similarity between the two major dietary patterns produced by both dietary assessment techniques. Nevertheless, a number of foods did not correlate between dietary patterns, which indicates differences in the dietary assessment methods, the 24 h MPR being acute and the FFQ habitual, and potential problems with the validity of the FFQ.

There are a limited number of studies evaluating the validity of dietary pattern analysis (Hu et al., 1999; Khani et al., 2004; Okubo et al., 2010; Nanri et al., 2012; Mills et al., 2015). One validation study on the dietary patterns of a sample of 127 men using an FFQ and two 1-week diet records reported that the major dietary patterns (‘Prudent’ and ‘Western’) defined by PCA from both dietary assessment techniques were correlated (Hu, 1999); suggesting that the two x 1 week dietary records represented a habitual diet- similarly to the FFQ. Khani et al. (2004) produced similar findings when validating an FFQ with 4 x 7-d dietary records to produce dietary patterns among 129 Swedish women. Likewise, a study on Japanese adults (n = 184) used factor analysis to derive dietary patterns from a self-administered diet history questionnaire (for foods consumed during the previous month) and 4 x 4-day dietary records (one 4-day record during each season for 1 year) (Okubo et al., 2010). The study showed relatively similar direction (r = 0.45–0.69) and magnitude of factor loadings of food groups from each dietary assessment technique, supporting their validity (Okubo et al., 2010). In a more recent study, PCA defined three Japanese dietary patterns from two FFQ and two dietary records (14-day and 28-day); Prudent, Westernised, and Traditional (r = 0.32–0.63) (Nanri et al., 2012); suggesting acceptable validity of dietary patterns produced. An alternative study using an FFQ and twelve 24 h dietary recalls found fair to modest validity of dietary patterns derived by cluster analysis (Funtikova et al., 2015); however, cluster analysis is a less sophisticated technique than PCA and is not directly comparable to the current study.

The EHS identified some similar dietary patterns to the 24 h MPR methods in the current study, namely the ‘Western’ and ‘Prudent’ patterns; suggesting acceptable validity for major dietary patterns produced. This provides additional evidence with respect to PCA being an acceptable method to determine major dietary patterns in traditional dietary assessment techniques (24 h MPR and FFQ). Nevertheless, the accuracy of dietary intake from both dietary assessment techniques was limited by the self-report nature including bias and human error by the participants. In addition, the PCA method (Laerd Statistics, 2018) involved excluding food groups such as Indian takeaway due to low levels of consumption in the participant sample during the 3 x 24 h MPR. Therefore, in the PCA analysis of 24 h MPR data, further analysis justified the removal of other potato, Indian, kebab, English takeaway, savoury sauces, fish, alcohol and tea. Yet in the EHS, further analysis eliminated alcohol, tea and coffee. Therefore, the same food groups were not used in each analysis. The removal of these food groups may have influenced the differences in dietary patterns produced using alternative dietary assessment methods. To help eliminate the error due to the self-reported nature of these methods it was decided to investigate the use of metabolomic biomarkers linked to the consumption of specific foods from spot urine samples, to help validate dietary patterns from self-reported dietary surveys.

6.5.2 USE OF A BESPOKE HOME-URINE COLLECTION-KIT FOR METABOLOMICS RESEARCH

Spot urine samples can be readily collected by individuals, is non-invasive, with expected high participant compliance (Khamis et al., 2017), thus making it an attractive bio fluid for metabolome biomarker discovery. Being able to collect urine samples in a home environment is a useful method for research of free-living individuals and the development of a prototype home urine collection-kit was important. In the current study, a high percentage (96%) of participants successfully collected urine samples via the bespoke urine sample collection-kit (Fig. 6.4). Most participants strongly disagreed (85%) that collecting urine samples at home was difficult, suggesting that they found the method easy (Fig. 6.5). However, 15% found the collection of a urine sample difficult (Fig. 6.5). During the 24 h MPR a number of participants reported not being able to fill one or more Vacutest sample tubes using a transfer straw; a possible reason for the reported difficulty. Nevertheless, this was factored during the design process and was the reason for collection of two duplicate samples per urine sample. A recent study by the researchers from IBERS, AU received 100% compliance in collection of spot urine samples with the prototype home-urine collection-kit (Wilson et al., 2016).

Previous studies, investigating food exposure in urine involved participants collecting 24 h urine samples (excluding first morning void) in large 2 or 3-L containers up to the following morning (including the first morning void) (Lloyd et al., 2013; Andersen et al., 2014). The current technique uses spot urine samples- the first morning void-, which reduces participant burden, as there is no need for 24 h urine collection. It has been previously demonstrated that, spot urine collected, either just before bedtime or as a first morning void, had as much value for dietary exposure assessment as urines collected over a 24 h period (Beckmann et al., 2013). The current study results are encouraging and support the use of the bespoke urine sampling collection-kit in future studies as a cost-effective approach to community based urine sampling. They could also facilitate monitoring of dietary behaviours in large-scale surveys and intervention studies.

6.5.3 Metabolite fingerprints associated with takeaway and habitual foods

The aim of the current section of the study was to identify metabolite fingerprints and potential discriminatory biomarkers associated with takeaway/unhealthy foods because self-reported dietary intake (from FFQ and 24 h MPR) does not represent an objective unbiased assessment. The results show some significant differences in the metabolite fingerprints of high, low and non-consumers for Indian, fast food, Chinese, English and kebab (Table 6.7). For example, Indian takeaway food (Fig. 6.11) showed discrimination of different levels of consumption of key foods potentially linked to less healthy diets. The results showed that Indian (Fig. 6.11) or Chinese (Fig. 6.13) have distinct classes and thus provides an opportunity to search for putative markers. Moreover, it appears that there is good biomarker potential for kebab (Fig. 6.15), Indian (Fig. 6.11), English (Fig. 6.14) and Chinese (Fig. 6.13). Nevertheless, the strong variation between chemical fingerprints may have occurred due to co-consumed foods (Beckmann et al., 2013). Therefore, the 24 h MPR data was analysed to identify the types of takeaway foods and co-consumed foods for the high takeaway food consumers, to suggest possible markers. For example, the majority of high Chinese consumers consumed a main dish in a sweet sauce (e.g. sweet chilli chicken), egg fried rice or chips, and a similar starter (spring rolls or prawn toast) but also consumed coffee throughout the day. For English takeaway, all participants in the high consumption group consumed chips with salt and vinegar and 75% had battered fish, and most consumed SSBs. Therefore, it is challenging to decipher new signals for foods or food groups when they are co-consumed and masked by another food/food group. However, if co-consumed foods are identified and there is a strong relationship then it is possible to use a known metabolite marker as an indication of consumption of particular foods/food groups. For example, coffee consumption may be a marker for Chinese takeaway food consumption, as established in the data from the 24 h MPR.

For total takeaway foods according to the Tw values the grouping did not produce distinct clusters (Table 6.7). Despite poor discrimination between high and low consumption of takeaway food, there was a good trend in the data on DF1 between very high and non-consumption (Fig. 6.16). This was a positive result because the method involved subjectively grouping participants into consumption classes and comparing a vast fingerprint of data. The applied class structure classified the samples just below the thresholds implemented. The poor discrimination among all takeaway foods might be a reflection of how to define 'high' and 'low' which in the current study was data-driven; a split sample into none, low, high and very high dependant on the median values of consumption. Another plausible reason is due to the variation within the total takeaway food group and the sheer number of foods included. Previous research stated that food groupings with a high degree of variety and number of food components creates difficulty in detecting exposure to that food group (Lloyd et al., 2013); suggesting that there may have been too many co-consumed foods and ingredients in the total takeaway food group. However, a positive and novel finding from this figure (Fig. 6.16) was being able to link metabolite fingerprints to takeaway food consumption. The unsupervised PCA method revealed natural clustering for Indian takeaway food consumption (Fig. 6.10); displaying three distinct clusters. The unsupervised results revealed similar findings for fast food, Chinese, English and kebab, inferring a good degree of variance (Appendix 9.42; 9.43; 9.44; 9.45).

Overall PC-LDA discriminants function analysis showed successful clustering using class structures for (supervised) high, low, none, indicating discrimination between the metabolite fingerprints of these groups of individuals.

This positive finding supports the use of this type of analysis for future studies on habitual consumers of a variety of foods including takeaway food. It may also lead the way in terms of validating dietary survey work in the future.

6.5.4 Target foods/metabolite signal annotation

The aim was to determine if the urine samples validate/reflect participant dietary intake from 24 h MPR and to validate known food metabolites or novel biomarkers. For all foods, distinctiveness and consumption-frequency range influenced the likelihood that differential dietary exposure could be detected. Supervised multivariate modelling output statistics indicated foods for which biomarker lead discovery was feasible. Results determined that the chemical composition of urine was related to the specific differences in the consumption levels of some key foods groups including poultry, coffee and alcohol. One of the top ranked signals causing the discrimination between high and no consumption of poultry was 3- Methylhistidine, appearing on the top ranked list in multiple adducts and isotopes (Table 6.8; Fig. 6.17). Several studies have reported an increase in urinary 3-methylhistidine after meat or more specifically fish/poultry consumption (Cross et al., 2011; Lloyd et al., 2011). Poultry was consumed in many formulations in the participant sample, in home-cooked dishes (fried chicken thighs, roast chicken, boiled chicken breast etc.) and takeaway meals (fried chicken, chicken Balti, sweet chilli chicken etc.). Poultry had a high positive loading in the ‘Takeaway Food' pattern and thus could be a marker for takeaway food consumption together with other more specific markers of takeaway foods.

A top ranked signal causing the discrimination between high and low or no consumption of coffee in the negative mode was Quinic acid, appearing as several adducts (Table 6.9; Fig. 6.18). Quinic acid is a sugar acid and is also a cyclitol, a cyclic polyol. More specifically, quinic acid is a crystalline acid obtained from cinchona bark, coffee beans, tobacco leaves, carrot leaves, apples, peaches, pears, plums, vegetables etc. Quinic acid is implicated in the perceived acidity of coffee (Esteban-Díez et al., 2004) and is a Chlorogenic acid breakdown product (plentiful in coffee). Trigonelline (N-methyl nicotinate) was also highly ranked in positive ionisation mode data as several different adducts and isotopes. Trigonelline is a well-published biomarker for the consumption of coffee (Lang et al., 2011; Lang et al., 2013; Rothwell et al., 2014). Both signals discriminated between the different levels of coffee consumption, highlighting the importance of using both ionisation modes. Coffee had a high factor loading in the Western dietary pattern and Cosmopolitan dietary pattern. Thus, it may be a marker for convenience style foods (buns, cakes and pastries) or healthier foods (eggs, fruit, wholegrains). Coffee had the highest negative factor loading for the Takeaway food dietary pattern, suggesting that coffee consumption is not linked to takeaway food consumption.

A top ranked signal causing the discrimination between high and low or no consumption of alcohol (wine and beer), Beer and wine was Ethyl glucuronide (Table 6.10; Fig. 6.19). Ethyl glucuronide is a natural human metabolite of Ethanol generated in the liver by uridine 5'-diphospho-glucuonyltransferase. Ethyl glucuronide is a well-published biomarker for the consumption of alcohol (Vázquez-Fresno et al., 2015). One of the top ranked signals causing the discrimination between high and low or no consumption of wine, but did not appear discriminatory for general alcohol or beer is Tartaric acid (Table 6.11; Fig. 6.20). The dietary origin of tartaric acid as natural organic acid in grapes at high concentrations is well-known. Tartaric acid has been demonstrated as a wine or grape/grape juice marker (Heinzmann et al., 2012; Vazquez-Fresno et al., 2012; Regueiro et al., 2014; Vázquez-Fresno et al., 2015; Garcia-Perez et al., 2016). Alcohol was consumed at varying amounts in the participant sample but was not correlated with any particular dietary pattern.

6.5.5 Dietary patterns and metabolite fingerprinting via non targeted MS

Urine samples from participants scoring high on each of the five 24 h MPR dietary patterns were analysed to determine if metabolite fingerprints could be used to classify these dietary patterns. There did appear to be discrimination between classes 1 and 2 and between 1 and 3 (Table 6.13). The three dietary patterns causing most discrimination were the Western dietary pattern, the Cosmopolitan pattern and the Takeaway Food pattern. All three patterns included many carbohydrate/sugary food items (Appendix 9.39), which mean that when broken down in the body and excreted in urine would make it difficult to differentiate patterns from one another. Further inspection of the foods consumed in these patterns suggested that the metabolites for cocoa, coffee, oils and fruit and vegetables might have caused some of the variance among clusters (Fig. 6.21). Cocoa was a key food found to be causing one of the discriminatory signals between classes; 3 and 7-Methylxanthine both published biomarkers of cocoa exposure (Llorach et al., 2010; Garcia-Aloy et al., 2015). Chocolate was identified in the Western dietary pattern from the 24 h MPR data which concords with the metabolite fingerprinting. Trigonelline appeared to be discriminatory between metabolite fingerprints: identified as a well-published biomarker for the consumption of coffee (Lang et al., 2011; Lang et al., 2013; Rothwell et al., 2014). Coffee was a key component in both the 'Western' and 'Cosmopolitan' dietary patterns.

Another key component causing discrimination between metabolite fingerprints was virgin olive oil. Tyrosol is a phenolic compound present in two of the traditional components of the Mediterranean diet: wine and virgin olive oil. It has been shown that urinary tyrosol concentrations are responsive to short-term dietary intake of virgin olive oil (Miró Casas et al., 2001). Oils and dairy free spreads were identified in the 'Western' and 'Takeaway' dietary patterns from the 24 h MPR data. Finally, a general fruit and vegetable marker was identified from the metabolite fingerprints and may represent foods consumed in the 'Cosmopolitan' dietary pattern. It has been indicated that Hippuric acid represents a useful biomarker for fruit, vegetable and fruit juice consumption (Krupp et al., 2012). The results show some validation of the dietary patterns of the study sample, from 24 h MPR data with metabolite fingerprinting. A recent study by Acar et al. (2019) assessed the effect of the new Nordic diet (NND) versus average Danish diet (ADD) on metabolic profiles and was able to identify potential biomarkers of intake of certain foods and biomarker signatures predictive of dietary patterns.

The findings from the EHS (FFQ) were unclear compared to those from 24 h MPR. This was because dietary pattern 1 accounted for most food items making it difficult to differentiate between other patterns (Appendix 9.40). There appeared to be some discriminatory signals from dietary pattern 1 (Fig. 6.27; 6.28; 6.29) which could potentially be unhealthy markers in line with previous work (Lloyd, personal communication). The majority of foods consumed in dietary pattern 1 were energy-dense nutrient-poor (e.g. cakes pastry buns, fried potato, high salt snacks, sweet sauces, sweets and sugar, SSB etc.) and included takeaway foods (Indian, English, fast food and Chinese) therefore supporting the concept of ‘unhealthy’ markers (Appendix 9.40). However, there are few published markers on these type of foods signifying that further work is required to confirm the structure of these putative unhealthy biomarkers.

6.6 Strengths and Limitations

This study had a number of strengths, including a good sample size for this type of research. The inclusion of human biofluid as an objective measure also strengthened the other dietary assessment tools; to aid measurement of dietary intake and assess validity of dietary assessment tools. This was a real world example of food intake and thus results are for habitual diet and incorporate all of the variation within a person’s diet and lifestyle as opposed to the controlled conditions in a laboratory. Previous research has shown more metabolites identified in urine (n = 154) than in serum (n = 39) were associated uniquely with one food (Playdon et al., 2016); supporting the use of urine in the current study. The use of the bespoke home urine collection-kit facilitated the recruitment of a large sample for metabolomics research and allowed participants to collect their sample at a time and place (i.e. home) that was convenient for them.

That said there were also a number of study limitations including both dietary assessment methods (24 h MPR and FFQ) were retrospective and prone to similar limitations, e.g. recall bias and misreporting (Chapter 4 and 5). The 24 h MPR were dependent on respondent's memory and there was a possibility of recall bias where individuals selectively recall food items (as with all self-reported methods). The repeat 24 h MPR were also time consuming and expensive to administer due to high interview burden. However, telephone interviews reduced cost and time. There was a degree of subjectivity in determining food groups before application of PCA, the treatment of the input variables and labelling of dietary patterns. Data-driven methods such as PCA may not derive comparable patterns in different populations. The proportion of consumers during the 3 x 24 h MPR was very low for some key food groups and could not be included in the PCA for dietary pattern analysis. A similar issue was reported by Batis et al. (2016).

There were also a number of limitations in relation to the metabolomics research. The sample population was balanced by age but unbalanced in gender (Table 6.3), with more females participating than males. Previous metabolomic analyses have noted gender specific metabolites, so where possible, when selecting high and low consumers of takeaways, a balanced group was selected and maintained. Samples were collected using a bespoke home collection-kit technique, therefore, guidance was restricted and some participants experienced more difficulty than in studies in laboratories with trained researchers to provide instructions. However, the acceptability questionnaire results showed that participants were reasonably happy. Urine sample deterioration may have occurred depending on time kept in fridge, temporary −20°C freezer and −80°C freezer (Scalbert et al., 2009). Some participants did not refrigerate samples, other samples spent prolonged durations in the post meaning further deterioration may have occurred prior to their arrival at LJMU (Khamis et al., 2017). Samples may have deteriorated at different speeds dependent on their specific gravity. The Vacustest tubes with caps removed and filled with urine by hand may have been less stable than those using the provided transfer straws. During the second transfer of frozen samples to AU, severe weather conditions caused the courier to retain the samples at a depot for an extra 5 days, during this time the samples were repacked on dry ice and there was a chance that the samples may have deteriorated during this time. Nevertheless, there was no evident deterioration in the samples using metabolomics fingerprinting. The presence and concentration of food biomarkers in the morning void urine samples would have been modulated by the timing of dietary intake. Some chemicals are excreted straight away and do not undergo any biotransformations and are not acted on by the gut microbiota. Whereas other are more habitual markers and are highly metabolised. Due to smaller sample size of 151 (49 less than the 200 target) some of the target groups for metabolomics analysis had < 8 participants which may have skewed the data slightly.

Due to the staggered design of the studies, some participants may have completed the EHS (FFQ) up to 8 months prior to completing the 24 h MPR and urine sample collection, thus there was a risk of dietary change over this period of time. Other factors that may have caused a change in dietary habits included season and illness. However, all efforts were made to record these limitations during each 24 h MPR and in the acceptability questionnaire. There was also a limitation due to the determination of ‘low’, ‘high’ or ‘very high’ levels of takeaway food consumption, which may have influenced the levels of discrimination during FIE-HRMS analysis and clustering of signals. If different cut off points were utilised the level of discrimination may have improved. This is of particular importance for future studies aiming to identify signals linked to unknown metabolites and where further testing and development may aid accuracy. The poor discrimination in some metabolite fingerprints may also be a reflection on the limitations of self-reporting/dietary survey. Nevertheless, with more work in this area in the future, urine metabolite fingerprints linked to diet studies and more standardised methods might be developed.

6.7 Conclusion

To conclude, the EHS was an acceptable tool to produce dietary patterns using PCA from population dietary consumption. The results from the 24 h MPR validated the major dietary patterns, namely the 'Western' and 'Prudent' dietary patterns produced from the EHS data. Dietary patterns from PCA on 24 h MPR data of the participant sample produced some similar patterns in the urine samples, specifically foods identified in the 'Western', 'Cosmopolitan' and 'Takeaway' dietary patterns, furthering validation of dietary assessment methods.

This study provided confirmation that urine samples can be collected in the home by participants with high compliance and acceptable ease of use. Metabolite fingerprinting on low, high and very high takeaway food consumer groups also showed a significant difference between patterns of consumption validating the self-reported dietary intake data. Very few diet studies on dietary patterns have included this level of validation thus, this research is pioneering in a number of ways. The results suggest that there is good biomarker potential for kebab, Indian, English and Chinese takeaway food. It has also been shown that the non-targeted metabolite fingerprinting can efficiently highlight metabolites associated with specific dietary components including poultry, coffee, alcohol, cocoa, oils and fruit and vegetables. The majority of the metabolites found in the current study are well-published biomarkers of specific food exposure (Lloyd et al., 2013). Nevertheless, some potential novel unhealthy markers were also highlighted but further work is required to confirm the structure of these putative biomarkers. High throughput FIE-HRMS fingerprinting provides a rapid and objective method to link urinary signals to dietary components recorded in 24 h MPR data. Non-targeted metabolomics such as flow infusion-high resolution fingerprinting could be used to validate results from other dietary assessment techniques to assess food consumption and could be linked to nutritional quality of dietary patterns.

This was an uncontrolled habitual population whereby all foods consumed and lifestyle choices varied among the group. This level of variation makes the research challenging but of high importance to study free-living individuals. This research is in its infancy and thus future studies are necessary to continue to develop, evaluate, and further expand metabolomics as a form of aiding dietary assessment (Subar et al., 2015). Recent advances in the omics field could help to further establish the list of reliable nutrition biomarkers (Naska et al., 2017) to produce an extensive biomarker panel for future research. Furthermore, metabolomics has great potential in terms of enhancing our understanding of the metabolic effects of certain foods/diets and could be key in developing future evidence based dietary recommendations (Brennan, 2019).

Chapter 7
Synthesis

7 SYNTHESIS

7.1 RECAP OF THESIS

The studies presented in this thesis contribute to a holistic overview of takeaway food consumers. It has addressed some of the gaps in the literature in relation to consumption of specific takeaway foods and wider dietary patterns in relation to socio-demographic and lifestyle factors. Moreover, it has built on a novel metabolite fingerprinting technique that distinguishes food exposure from urinary samples. This project included three main stages of data collection. The initial stage (Study 1, Pilot) included the simultaneous creation, testing and optimisation of the 'Eating Habits Survey' (EHS), a modified version of the EPIC-Norfolk food frequency questionnaire (FFQ), designed to analyse habitual and takeaway food intake, along with background information. Similarly, the 24 hour 'Multiple Pass' dietary recalls (24 h MPR) dietary assessment technique was tested and piloted for the main study. Both methods also provided the opportunity to gather preliminary data to experiment with the principal component analysis (PCA) technique to determine dietary patterns.

The second (Study 2) utilised the tested EHS for a cross-sectional observational study of 1724 adults (aged 18-64 years) in Merseyside, UK. Consumption of 212 habitual and takeaway foods and socio-demographic (including age, education level, ethnicity etc.) and lifestyle factors (physical activity, cigarette smoking and alcohol consumption etc.) were self-reported. PCA was used to determine dietary patterns of the population and associations were investigated between takeaway food intake, wider dietary patterns and socio-demographic and lifestyle factors. The second study also allowed for recruitment of a sub-sample of participants for the final study.

The third (Study 3) investigated the dietary patterns of 151 adult takeaway food consumers using the EHS and 3 x 24 h MPR. A group of participants did not consume takeaway foods during the study period and these were labelled non-consumers for analysis purposes, however, all participants had reported having consumed takeaway food within the previous 12 months (inclusion criteria for Study 2). Dietary patterns were identified through PCA using both dietary assessment techniques. Additionally, after each 24 h MPR participants collected spot, first morning void, urine samples using a bespoke home urine collection-kit. Metabolite fingerprints were created from urine samples using flow infusion electrospray (FIE) ionisation high resolution (HR) mass spectrometry (MS) and were analysed with machine learning data techniques, including Random forest (Beckmann et al., 2013; Beckmann et al., 2016). Next, Ultra High Performance Liquid Chromatography-High Resolution MS (UHPLC-HRMS) and Tandem mass spectrometry (MSn) was used for structural identification of putative biomarkers. Urinary metabolite fingerprints were then subsequently compared with the consumption of takeaway foods, habitual foods and wider dietary patterns.

The formation of the research collaboration with IBERS at Aberystwyth University was highly significant and involved ascertaining internal and external funding, and multi-site organisation. The collaborative project also required an amendment to the original study design, which included the removal of a qualitative study (one to one semi-structured interviews), to maintain a realistic time-scale. The combined use of two dietary assessment methods with supplemental biomarker discovery via metabolomics allowed for several layers of dietary analysis and validation. Thus creating a detailed and innovative study on takeaway food consumers.

7.2 DISCUSSION OF FINDINGS

7.2.1 DEVELOPMENT OF A MODIFIED FFQ TO MEASURE TAKEAWAY AND HABITUAL FOOD CONSUMPTION IN RELATION TO BACKGROUND CHARACTERISTICS. IN ADDITION, TO TEST DIETARY PATTERN ANALYSIS FROM FFQ AND 24 H MPR.

Results from each of the studies have been previously published (Publications list, Page 22-23) (Janssen et al., 2018a; Janssen et al., 2018b; Janssen et al., 2018c; Lloyd et al., In Press). One of the main purposes for carrying out the Pilot Study was to create a suitable dietary assessment tool to measure both takeaway food and habitual food consumption and to test dietary pattern analysis. This crucial development stage took place between January and April 2016 and encompassed a sample size of 26 LJMU university staff and students. The findings of Study 1 showed that 23% of participants in the pilot consumed takeaway food once per week or more (Fig. 4.1), lower than the average reported amount of 27% in the UK (Food Standards Agency, 2014; Public Health England, 2017a). In this study, PCA was recognised as an appropriate technique for dietary pattern analysis, generating three distinct dietary patterns; 'Traditional, 'Cosmopolitan' and 'Convenience' (Fig. 4.6). The three-component solution explained 42.9% of the variance in dietary intake. Interestingly, the elements of the diet that tracked together in the Convenience dietary pattern included energy-dense nutrient-poor habitual foods (refined grains, cakes, pastries etc.) and takeaway food (Chinese, Indian, kebabs, fast food etc.). Thus, the findings corroborates the ideas of Poti et al. (2014), who suggested that fast food was a marker of an unhealthy Western dietary pattern in children. In support of this concept, Mueller et al. (2018) reported that young adults never eating meals away from home was associated with higher adherence to the Prudent and lower adherence to the Western and Alcohol patterns. Overall, the dietary patterns of the sample supports previous research on the use of PCA to produce dietary patterns (Thorpe et al., 2016; Mertens et al., 2017; Roberts et al., 2018). However, with a small sample size, the findings of Study 1 might not be generalised to wider populations. The use of the EHS and 24 h MPR in combination provided long-term and short-term assessment of dietary intake that were used for validation purposes; the foods consumed during the 24 h MPR validated dietary patterns identified from the EHS data (Fig. 4.7). This finding, whilst preliminary, suggested that 24 h MPR was a suitable method for validating dietary patterns from FFQ data. The results from the pilot (Study 1) were encouraging and supported the opening of the main study (Study 2).

7.2.2 Habitual and takeaway food consumption, socio-demographic and lifestyle characteristics and wider dietary patterns in a Merseyside community sample.

Study 2, repeated some of the methods developed and tested during the pilot study; however, this time on a larger population in Merseyside. The initial finding was that takeaway food consumption was higher in the participant sample than in the previous research (Food Standards Agency, 2014; Public Health England, 2017a), as expected due to the inclusion criteria (consumption of takeaway or fast food at least once in the last year). The majority (46%) of the sample consumed takeaway food 1-3 times per month and over one third (37%) consumed takeaway food at least once per week (Fig. 5.2), similar to that of consumption rates in Australia (Miura et al., 2012). Having more children in the household, a physically active occupation, participating in moderate and vigorous physical activity, frequent smoking, increased alcohol consumption, poorer health status, and increased BMI were positively correlated with takeaway food consumption ($p < 0.005$) (Table 5.5). Increased age and a higher education level were negatively correlated with takeaway food consumption ($p < 0.0005$) (Table 5.5). The socio-demographic and lifestyle factors as shown above correlate to varying degrees with takeaway food consumption, which agrees with previous research (Lachat et al., 2012; Adams et al., 2015). Furthermore, in line with preceding literature, takeaway food consumption was overall associated with lower socio-economic status (measured by education and occupation) (Miura et al., 2012; Barton et al., 2015; Goffe et al., 2017), male gender (Smith et al., 2009; Orfanos et al., 2009; Food Standards Agency, 2014) and younger age (Smith et al., 2014; Adams et al., 2015; Liu et al., 2015; Athens et al., 2016). This study demonstrated, for the first time, that specific takeaway food types (Indian, Chinese, fast food etc.) are consumed in contrasting ways based on population characteristics. There was also a correlation between fast food consumption and smoking, and Indian takeaway food with alcohol (Table 5.6). Indian takeaway food was an anomaly for many associations; it was consumed heterogeneously among the sample with a slight increase in individuals with higher educational attainment (Table 5.6). These significant findings, yet preliminary, could help inform policy and interventions to target the impacts of takeaway food on obesity and related non-communicable disease, by better understanding how takeaway food consumption links to other factors in adult lives. Moreover, the current findings add to a growing body of literature on the determinants of takeaway food consumption.

Four dietary patterns were derived from the PCA based on scree plots and eigenvalues and labelled according to the factor loadings and previous literature; the 'Western' dietary pattern; the 'Prudent' pattern; the 'Takeaway Food' pattern and the 'Meat and Dairy' pattern (Fig. 5.12). Most of the dietary patterns produced in each study contained foods considered as ultra-processed shown to be harmful to health (Monteiro et al., 2011; Fiolet et al., 2018), thus suggesting that the majority of the participant sample had poor dietary habits. Takeaway foods were split by type in the current study for dietary pattern analysis and thus the food groups cannot be directly compared to those from Study 1, which analysed total takeaway food consumption due to limited participants. However, an important finding in Study 2 was that all of the individual takeaway food groups (Chinese, English, fast food, kebab, pizza and Indian) correlated with one another, along with pizza and sausages (Fig. 5.12) to produce one dietary pattern; suggesting that those consuming takeaways consumed them frequently and with other convenience foods. There were no previous studies to directly compare results that analysed dietary patterns by separating takeaway foods.

Having children in the household and poorer health status was associated with increased consumption of the 'Western' dietary pattern. Whereas older adults, higher educational status, increased alcohol consumption, and moderate/vigorous exercise were positively associated with the 'Prudent' dietary pattern and negatively associated with the 'Western' pattern (Table 5.11). The 'Takeaway Food' dietary pattern was associated with increased adults in the household, poorer health status, smoking, increased alcohol consumption, increased physical daily occupational activity and moderate/vigorous exercise (Table 5.11). Lastly, the 'Meat and Dairy' dietary pattern was positively ($p < 0.005$) associated with older age, more children in the household, poorer health status, smoking, increased BMI, lower education and increased daily occupational activity (Table 5.11).

This study has gone some way towards enhancing our understanding of takeaway food consumption and links to wider dietary patterns. Along with the socio-demographic and lifestyle determinants of the foods consumed. The need for a greater understanding of takeaway food consumption and wider dietary patterns in the UK is emphasised by the fact that the majority of the study sample consumed a significant amount of 'unhealthy' foods. Thus, the evidence base needs to be coherent to support strategies to improve public health. This supports the continual collection of dietary intake data of the population, such as the NDNS data, that can be used to inform nutrition policy and assess diet-disease associations (Subar et al., 2015). Such population based surveys could be improved by including further detail on takeaway and convenience food; especially as they are increasingly being consumed.

7.2.3 The dietary patterns of a sub-sample of data-driven takeaway food consumers and validation of dietary assessment techniques using metabolomics. In addition, the acceptability of a home urine

In Study 3 PCA produced five dietary patterns from the 24 h MPR, these were classified as; 1) Western, 2) Cosmopolitan 3) Takeaway 4) Prudent and 5) Sweet (Fig. 6.9), and four patterns from the EHS, classified as; 1) Western, 2) Snacks, 3) Healthier and 4) Takeaways and Meat (Fig. 6.8). The EHS was determined as an acceptable tool to produce dietary patterns using PCA with the results from the 24 h MPR validating the major dietary patterns; 'Western' and 'Prudent'. Study 3 provides additional evidence with respect to determining dietary patterns via PCA for participants in the UK and using 24 h MPR to validate FFQ dietary data. These findings corroborate those discussed previously (Hu, 1999; Khani et al., 2004; Okubo et al., 2010; Nanri et al., 2012) who also validated FFQ dietary patterns with dietary records.

The third study makes several noteworthy contributions to the field of metabolomics on the use of urine collection in the home to measure food exposure in a free-living population. Firstly, a bespoke home urine collection-kit was developed by researcher from IBERS, AU, and this prototype collection-kit was used and evaluated by participants in the study. Overall, the majority (96%) of participants were able to successfully collect urine according to the provided photographic instructions (Fig. 6.4), however, 15% found the collection of a urine sample difficult (Fig. 6.5). Feedback on the difficulty of collection has been reported back to the developers for fine-tuning of the bespoke kit. Using FIE-HRMS coupled with machine learning, this study, for the first time, has determined differences among the metabolite fingerprints of takeaway food consumers showing that there is good biomarker potential for kebab, Indian, English and Chinese takeaway food (Table 6.7). UHPLC-HRMS and MSn allowed further structural identification of biomarkers associated with takeaway food consumption including markers for poultry (Fig. 6.17), coffee (Fig. 6.18) and alcohol (Fig. 6.19). In addition, urinary biomarkers associated with the major dietary patterns from 24 h MPR data were identified and validated against previous research.

The present study adds to the literature, for the first time, evidence of metabolite fingerprinting for takeaway food exposure. This highly novel area of research requires further development to identify the best technique to link dietary surveys to untargeted and targeted metabolite profiles. Hence, it would be advantageous to progress, evaluate, and further expand such technological methods of dietary assessment, including dietary biomarkers (Subar et al., 2015). In due course, urine biomarkers indicative of exposure to foods ranging from healthy to unhealthy could be integrated into a diagnostic population screening method in order to objectively measure food exposure. Moreover, it is predicted that putative biomarkers could replace some traditional dietary assessment methods in the future due to their translation into practical measurements of dietary exposure (Beckmann et al., 2013).

7.3 POTENTIAL OUTCOMES OF THIS RESEARCH AND IMPLICATIONS FOR TAKEAWAY FOOD CONSUMERS

The use of dietary patterns could help to improve adherence to dietary guidelines and reduce the consumption of unhealthy foods. Instead of focusing on macro- and micronutrients that are not meaningful to the general population, dietary patterns are a viable system for public health practice (Tapsell et al., 2016). Moreover, now that the burdens of disease have shifted from communicable to non- or socially-communicable in many countries (Popkin et al., 2012; Moreira et al., 2015; Clark, 2017), the focus also needs to shift from nutrients to dietary patterns. Concerning takeaway food consumption, this study has evidenced that those consuming high quantities of takeaway foods have poor habitual dietary patterns, a major problem for obesity and poor health outcomes. This trend supports previous literature (Poti et al., 2014; Zheng et al., 2016) and provides sufficient rationale to implement interventions and strategies to help improve the foods consumed; particularly for males, younger age groups, those from lower socio-economic status. A multi-pronged approach needs to be encouraged at a local and national policy level to combat the impacts of takeaway food. At the local level, additional planning policies to promote healthier food and drink, restrict the opening of new takeaway food outlets, and working with schools would be advantageous. At the national level, reformulating products (sugar and salt reduction), taxation on sugar and junk foods, plus marketing campaigns have the potential to contribute to a healthier nation. Reducing portion size and reformulating takeaway foods to improve their nutritional composition may significantly decrease consumption of high-energy foods (Ziauddeen et al., 2015). Public health policies that simply involve providing information are unlikely to be effective (Hillier-Brown et al., 2017). For these types of initiatives to be of success, they will need a multi-agency approach with co-partners from across the public, private and voluntary sectors. For example, Lake et al. (2017) suggest formal and soft approaches to improve collaborative working between planners and public health to address obesity such as written responsibilities, aligning spatial and health priorities and providing planners with public health leadership roles.

7.4 STRENGTHS AND LIMITATIONS

A detailed account of the strengths and limitations for each study can be found prior to the conclusion in each study chapter. The key strengths for the project are that methods were piloted (Chapter 4) and showed that PCA was an appropriate technique for dietary pattern analysis, generating three distinct dietary patterns. Study 2 (Chapter 5) was a population-based study with a large sample size of 1724 participants and included the collection of multiple-layers of dietary assessment data to validate findings. Takeaway food consumption was analysed in more detail compared with previous research by splitting into several specific takeaway food categories and studied across socio-demographic groups and for individual lifestyle factors. The inclusion of human biofluid in Study 3 (Chapter 6) as an objective measure also strengthened the other dietary assessment tools; to aid measurement of dietary intake and assess validity of dietary assessment tools. The development of the bespoke home urine collection-kit also allowed participants to collect their urine sample at a time and place (i.e. home) that was convenient for them. This was a study of free-living individuals; particularly important when analysing habitual diet consumption/exposure, as opposed to controlled conditions in a laboratory.

Some of the major limitations from the project are that the results for the participant sample in each of the studies may not be entirely representative of adults in Merseyside, adults in the UK or further afield. Yet, the dietary patterns resemble those described in previous UK studies suggesting a level of homogeneity. Convenience and snowball sampling techniques means that participants self-selected, and with no empirical information on non-participants bias cannot be excluded. There was an unknown degree of misreporting from self-reported dietary intake and lifestyle information. However, the inclusion of an objective biomarker to counterbalance this has shown some degree of acceptability in terms of validating the 24 h MPR method for dietary assessment. In addition, the 24 h MPR also validated major dietary patterns produced from the EHS developed during this research. Metabolomics was used for the first time to research takeaway food metabolite fingerprints and despite limitations (low sample size in some groups, many co-consumed foods, self-reported dietary intake, the arbitrary classification of low – high levels of takeaway food consumption) the results showed good biomarker potential for many of the takeaway food groups consumed.

7.5 RECOMMENDATIONS FOR FURTHER RESEARCH

7.5.1 EXPLAIN THE GEOGRAPHICAL MAPPING OF THE TAKEAWAY FOODSCAPE

Many of the neighbourhoods, towns, and cities in the UK, particularly those that are deprived, are described as obesogenic environments; where food choices are limited to unhealthier options and encourage excess weight gain and obesity (Public Health England, 2017a). It has also been reported that takeaway food outlets often cluster near UK schools, with children from deprived neighbourhoods consuming more takeaways and displaying higher obesity rates than those in less deprived neighbourhoods (Turbutt et al., 2018). Reducing obesity, particularly among children, is a national and international priority (World Health Organization, 2012; Public Health England, 2017a). The current study initially planned to include data from the Liverpool local authority on number and types of takeaway food outlets to illustrate density of takeaway food outlets in defined geographical locations. This would involve mapping of the local takeaway foodscape using geographical information systems to explain how the physical food environment shapes food choices. The collaborative metabolomics research project with IBERS, AU replaced this planned section of the study and due to the limited timeframe to complete the research was not addressed. However, there is still a rationale to complete this additional research.

Postcode data of the participant sample from Study 2 means that this research is possible without the need for further recruitment or data collection. Since the original planned use of the local authority records, the CEDAR food environment assessment tool (UKCRC Centre for Diet and Activity Research (CEDAR), 2017b) was developed and could supply supplementary data regarding takeaway food density in the current study area. The combined use of this data would allow for comparisons to be made between the location of takeaway food consumers and their local takeaway foodscape to explain the ways in which various demographics interact with their physical food environment. This research may highlight populations that need the most attention from locally focused public health strategies. There is the potential to expand the current research project nationwide; research on takeaway food consumption and wider dietary patterns in other areas of the UK could provide further evidence to support future policy and planning to improve public nutrition and health.

7.5.2 ADVANCE THE STATISTICAL ANALYSIS OF DIETARY PATTERNS

This research project was influenced by the takeaway or 'fast food' literature along with nutrition related papers on dietary pattern analysis. PCA has been widely used within this area of research however, previous studies may not be produced with expertise in statistics or the nutrition data to develop the most appropriate methods to produce dietary patterns. Therefore, there is the potential to produce a firmer methodological framework for performing dietary pattern analysis which could help standardise the analysis for future research. There was some difficulty when deciding on the most appropriate technique to produce dietary patterns in this study as there needs to be a stronger theoretical basis of how dietary patterns are defined and developed. Like with all quantitative research, the dietary pattern approach requires rigour and thus the next step would be to collaborate with a statistician or data scientist to re-analyse the data using other statistical techniques. There is an opportunity to do triangulation with the collected data (FFQ, 24 h MPR and Metabolomics). Furthermore, in nutrition literature dietary quality scores and diet index have been used (Reedy et al., 2014; Mercille et al., 2016) to assess the quality of individual's and population's diets and could be applied to the data from this study.

7.5.3 ASSESS THE IMPACT OF TAKEAWAY FOOD AND WIDER DIETARY PATTERNS ON HEALTH

As discussed in the earlier literature, it has been reported that regular takeaway food consumption can negatively affect coronary heart disease, type 2 diabetes and obesity risk markers in children (Donin et al., 2017) and adults (Bahadoran et al., 2012; Smith et al., 2012). Moreover, dietary patterns similar to a western dietary pattern have been associated with increased CVD and coronary heart disease risk, and type 2 diabetes (Maghsoudi et al., 2015; Rodriguez-Monforte et al., 2015; Al Thani et al., 2016; Atkins et al., 2016; Mertens et al., 2017). The current study has identified dietary patterns and linked to urinary metabolite profile/patterns, therefore the next step could be to investigate takeaway dietary patterns and measurable disease risk markers in the urinary metabolites from the collected urine samples (Chan et al., 2016). A recent study on US adults was the first to examine specific unhealthy foods with cardiometabolic risk factors considering ethnicity, sex and age-groups (Vaccaro Joan et al., 2018). Therefore, a fascinating area of research would be to include traditional CVD risk markers and a plasma/serum metabolomics approach in addition to the urine collected to assess the impact of takeaway foods and wider dietary patterns on public health. This research will help to ascertain if there is an association between takeaway food consumption, habitual dietary intake, and cardiovascular/cardiometabolic risk factors. Plasma/serum would strengthen the study by revealing additional markers, and include previously established ones.

It was previously reported that many UK takeaway meals were exceptionality high in portion size, energy density, total fat, salt, and sugar (Jaworowska et al., 2012; Jaworowska et al., 2014) and also excessively high in saturated fatty acids and some trans fatty acids (Davies et al., 2016) suggesting that regular consumption may contribute to a variety of non-communicable diseases. A review of the literature regarding takeaway foods and health outcomes concluded that findings were limited and recommended more studies to gain a stronger understanding of the nutrition and health consequences of takeaways food consumption (Jaworowska et al., 2013). Furthermore, gaining evidence of the risk factors associated with takeaway food and wider dietary patterns in the UK population may help to determine and support the most effective policies and strategies to reduce the negative impacts on public health.

An additional gap that the current study could address would be to assess nutrient intake linked to dietary patterns. Future plans for the data include the conversion of dietary survey data into nutrient intake using nutrition analysis software such as Dietplan 7 (Forestfield Software Ltd, West Sussex, UK) or MyFood24© (Dietary Assessment Ltd 2018, Leeds, UK). This would be specifically suitable for the final study (Study 3) given that the 24 h MPR is a more detailed measure of food consumption than an FFQ and metabolite fingerprinting was included.

7.5.4 INVESTIGATE TAKEAWAY FOOD MARKERS FOR METABOLOMICS ADVANCES

This study was the first to investigate biomarkers and patterns for takeaway foods in a cohort study (free-living individuals consuming their habitual diet), and has demonstrated that different metabolite patterns exist among takeaway food consumers. This has acted as a foundation for further research and it would be advantageous to continue the research by conducting a takeaway food feeding study that would include a number of specific takeaway food meals. Short-term intervention studies measure acute dietary exposure of specific foods and dietary components. Previous studies have included foods of high public health importance such as sucrose (Beckmann et al., 2016), wine (Regueiro et al., 2014), salmon, broccoli and raspberries (Lloyd et al., 2011) and chicken, red meat, processed meat, and fish (Cheung et al., 2017). However, there are currently no known intervention studies investigating takeaway foods.

As takeaway food is a cause for public health concern, it is warranted to study takeaway foods in detail through a short-term (4-week) food intervention study (possibly randomised cross-over or parallel feeding trial). This is to allow for specific takeaway foods to be targeted, for markers of CVD to be identified, and to provide more accurate measurement and assessment than in free-living individuals. Randomised clinical trials are considered the gold standard, however, whilst a crossover trial provides each participant with two or more sequential treatments acting as own control (Mills et al., 2009), the washout period for takeaway food may take some time. Nevertheless, parallel introduces other bias such as achieving like for like at baseline and requiring larger sample sizes.

The identification and validation of novel metabolites in urine associated with takeaway foods may have future potential as biomarkers of acute exposure to takeaway foods and thus could be added to the chemical libraries for the food metabolome (Scalbert et al., 2014), which would be of high public health importance. In general, incorporating metabolomics into forthcoming food and nutrition research studies has the potential to enhance understanding of how food impacts public health (Brennan, 2019).

7.5.5 A QUALITATIVE STUDY TO EXPLORE SELF-REPORTED FACTORS ASSOCIATED WITH TAKEAWAY FOOD CONSUMPTION AND WIDER DIETARY PATTERNS

The current research adopted a quantitative approach, with the aim to determine takeaway food consumption and wider dietary patterns of a Merseyside population by using principal component analysis (PCA). In addition, the research aimed to identify novel metabolite biomarkers associated with takeaway foods/unhealthy eating as a means of validation of self-reported food consumption. Therefore, this project outlined validity and reliability and acted as a base to initiate further research (Dawson, 2009) and did not provide rich insights into phenomena. The lack of existing quantitative research meant this study was appropriate for the field of study. That said, to further this research in terms of the factors influencing consumption, a qualitative study could provide explanatory description. Qualitative research attempts to widen and deepen understanding of how things in the social world came to be the way they are (Hancock et al., 2007).

There are currently a limited number of qualitative studies on the experiences of takeaway food consumers. A recent study used constructivist grounded theory methodology to explore the sociocultural experiences of takeaway food consumers in Rusholme, Manchester (Blow et al., 2019). The research produced three superordinate themes from thirteen interviews including social factors, personal factors and resources (Blow et al., 2019). A similar approach could be used to provide insight into why people consume takeaway food in the current study and their experiences of the local foodscape. A previous study, exploring the underlying drivers of eating out in Singapore used focus group discussions to find that the primary driver was the large variety of easily available and affordable hot cooked food, particularly at hawker centres (Naidoo et al., 2017). Caraher et al. (2014) also used focus groups to add further detail to observational data on school children's use of nearby fast food outlets. Focus group discussions or one-to-one interviews could be used for participants involved in the current study. The data collection during the 24 h MPR in the current research lends itself to a more explicit conversation (independent of use of the 24 h MPR method) to acquire a deeper understanding. This will provide an opportunity to gain insight into the influences on the foods consumed among the participants (takeaway and wider dietary patterns) along with their feelings, perceptions, and attitudes towards takeaway foods. This type of qualitative data could supply valuable information to support future interventions and strategies to meet consumer demands whilst being underpinned by a public health agenda.

7.5.6 Delivered restaurant food and 'Dark Kitchens' as an emerging area in a changing landscape of takeaway foods

There is a new emerging way in which consumers are accessing takeaway food in the UK, driven by technology (Richardson, 2019). Online food delivery companies such as Deliveroo and Uber Eats have made it possible to order takeaway food from almost any type of restaurant, including fast food from national/multinational fast food chains (such as McDonald's, Burger King, Pizza Hut and Kentucky Fried Chicken), meals from chain restaurants (such as Las Iguanas, Byron, Wagamama and Zizzi), food from independent local restaurants, and exclusives sold only through the delivery company (Deliveroo, 2019b; Uber Eats, 2019). In 2016, it was also reported that Deliveroo were selling a Michelin star menu from Galvin at Windows (Alexander, 2016). This dramatic change to the accessibility of food made to order and consume not only makes it harder to define what constitutes takeaway food but also limits understanding of the types of takeaway foods being consumed, and if the increased availability effects public health. Thus, making it a topical and fascinating area of research that requires further investigation.

In conjunction to the change in delivery style of takeaway foods, a number of news articles have reported a rapid rise in ‘dark kitchens’ or ‘pop-up kitchens’, set up specifically to produce takeaway foods for delivery, often in converted shipping containers located in parking lots and industrial plots (Ungoed-Thomas, 2019; Bradshaw, 2019). According to Deliveroo, their ‘Editions platform’ helps businesses segregate in-house orders to delivery orders, supports restaurant businesses to open up in other areas, and smaller businesses to start-up by setting up kitchens to produce food, without having the overheads of a high-street facing establishment (Deliveroo, 2019a; Cook, 2017). An inherent problem with ‘dark kitchens’ is that they are not registered as takeaway or restaurant to avoid complex planning permissions (BBC News, 2019). As a result, there have been concerns raised regarding planning permission and regulation, with some councils reporting a lack of planning applications and neighbours living nearby complaining about noise (Butler, 2017a; Butler, 2017b; Ungoed-Thomas, 2019). Therefore, due to the rapid development of this new platform, research is warranted to understand the effects of such food production on communities/neighbourhoods, the takeaway food industry (particularly small independent restaurants serving takeaway food), the nutritional composition of these foods and consumers themselves.

7.6 SUMMARY AND CONCLUSION

Food, whether takeaway food or home-made meals, is linked to all aspects of life and most importantly health, thus the food consumed will either keep individuals in good health or increase pressure on already exhausted health systems. The obesity situation being endemic in many of the World's countries, most notably those adopting modern "westernised" diets and lifestyles, suggests that today's diet contains an overabundance of energy-dense foods. The findings from this study adds to the literature on takeaway foods and wider dietary patterns and has helped to highlight a cause for concern with regards to the diets of many of those in this research project, which may reflect takeaway habits and wider dietary patterns of populations in other areas of the UK. Many of the dietary patterns identified in this study including the Western, Takeaway Food, and Meat and Dairy patterns comprised energy-dense nutrient-poor western type foods, with a minority consuming prudent or 'healthier' dietary patterns. Those consuming takeaway foods tended to have more western type dietary patterns, and correlated with other health harming behaviours such as frequent smoking and increased alcohol consumption.

The predominant recurring dietary patterns in Study 1, 2 and 3 produced by PCA were the Western and Prudent dietary patterns. The 24 h MPR validated a number of foods in the Western and Prudent patterns from EHS in Study 3 suggesting acceptable validity for major dietary patterns produced. Nevertheless, the three dietary patterns causing most discrimination in the urinary metabolite fingerprints were the Western dietary pattern, the Cosmopolitan pattern and the Takeaway Food pattern. The use of metabolomics in the current research has excelled the contribution made to the knowledge of takeaway foods. It has been noted that a key application of metabolomics in nutrition research is the identification of biomarkers of food intake that have the potential to act as objective biomarkers and help increase the accuracy of dietary intake data (Brennan, 2019). Findings from Study 3 not only helped to validate dietary intake data from two dietary assessment techniques (EHS and 24 h MPR) but also determined, for the first time, differences among the metabolite fingerprints of takeaway food consumers and has shown potential for novel biomarkers including kebab, Indian, English and Chinese takeaway food.

Out-of-home foods are a huge part of the economy in the UK; however, they are of great public health importance due to the potential adverse effects to health. With the recent report showing that UK high streets have the highest rates of takeaway food outlets seen in the last 8 years (Office for National Statistics, 2018) it is important for local authorities to have legislation in place to help drive forward an effective redesign of damaging neighbourhood food environments (Lake, 2018; Royal Society for Public Health, 2018). A collaborative approach to reducing the negative effects of takeaway food on public health would be preferential; including support and participation from the food industry, government, local authorities, and public health bodies. It is thought that by having a multi-agency approach that this may contribute to improvement of the obesity crisis (Townshend et al., 2015; Lake et al., 2017). The findings from this research should inform further research in this highly topical area, to contribute to a deeper level of understanding of takeaway food consumers and wider dietary patterns and to provide the evidence base to influence the formation of policy and interventions to ameliorate takeaway food consumption and provision locally and in other areas.

Chapter 8
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8 References

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Chapter 9
Appendices

9 APPENDICES

9.1 TABLE 1. SUMMARY OF STUDIES INVESTIGATING THE EFFECTS OF SOCIETAL INFLUENCES AND/OR INDIVIDUAL ACTIVITIES ON TAKEAWAY FOOD CONSUMPTION

Authors, type of study	Country	Study aim	Participants and setting	Methods	Results	Limitations
Welch et al. (2009) Cross-sectional	Australia	Examine the perception of time pressure as a barrier to healthy eating and physical activity	1,580 women (aged 18-70 years) from 45 neighbourhoods within approximately 25km of Melbourne's central business district	Self-reported frequency of dietary intake including questions on fast food intake, international Physical Activity Questionnaire	41% of the women sampled reported time pressure (due to long hours in work or study) as a barrier to healthy eating. The same individuals were more likely to consume fast food more often	Cross-sectional design precludes the ability to infer causal relationships, results may not be applicable to men or other populations, objective measures such as length of work were not included
Botonaki and Mattas (2010) Cross-sectional	Greece	Examine the way personal values are associated with convenience food consumption	729 adults (aged 18+ years) responsible for food purchasing and preparation, and not working in market research or advertising	Questionnaire survey and Schwartz theory of values; power, achievement, hedonism, stimulation, self-direction, universalism, benevolence, tradition, conformity and security	People with the orientation to consume convenience foods were associated with motivations to seek new experiences, act independently and enhance their own personal interests	Cross-sectional design precludes the ability to infer causal relationships, small study area (Thessaloniki in Greece) therefore results may not be generalized, food choice is often habitual and impulsive and not correlated with values
Maubach et al. (2009) Cross-sectional	New Zealand	Elicit factors influencing food purchases by parents and explore	15 parents (4 men, 11 women) who had children aged 5-12 years in their	Face to face semi-structured interviews about a typical	Pleasing their children, completing shopping quickly, selecting familiar brands and maintaining	Parents recruited from one geographic area in New Zealand and may not be generalized to other populations, participants in

		their understanding of nutrition labels	care in April/May 2007	shopping expedition with thematic analysis	routine all impacted use of nutrition labels	the study were literate whereas around half of New Zealand adults have low literacy levels
Stok et al. (2014) Cross-sectional	Poland, Portugal, UK and the Netherlands	Investigate associations of subjective peer norms with adolescents healthy and unhealthy food intake	2,764 European (pre-) adolescents (aged 10–17 years) from 24 schools in 4 countries	5-point Likert scale for subjective norms and healthy eating intentions, food intake measured as servings per day, Theory of Planned Behaviour	Subjective peer norms were associated with adolescents' healthy eating intentions and self-reported intake of healthy and unhealthy food	Cross-sectional design precludes the ability to infer causal relationships, self-reported food intake may not be reliable, findings may not be generalized to all adolescents
Lowry et al. (2015) Cross-sectional	US	Describe the association of sedentary behaviour and physical activity with dietary behaviours of US high school students	11,429 students (aged 14-17 years) from private and public high schools in 50 US states plus District of Columbia	Data from the 2010 National Youth Physical Activity and Nutrition Study	Physical activity behaviours and dietary behaviours are strongly related and do not vary by sex, race/ethnicity, grade, body weight status, or weight management goals of students	Cross-sectional design precludes the ability to infer causal relationships, youth from US high schools may not be representative of other individuals in this age group, self-reported data may incur under/over-reporting

9.2 Table 2. Summary of studies investigating the effects of the food environment and/or socio-economic differences on takeaway food intake

Authors, type of study	Country	Study aim	Participants and setting	Methods	Results	Limitations
Miura et al. (2012) Cross-sectional	Australia	Examine the socio-economic differences in types and frequency of	903 adults (aged 25-64 years) from Brisbane, Australia found on the electoral roll	Overall consumption and 22 specific takeaway foods measured using a FFQ*, takeaway foods grouped as healthy and less healthy and	The least educated participants were more likely to consume takeaway food > 4 times/month and made unhealthier food choices compared to their highly	Possible nutrient variation between healthy or unhealthy takeaway foods when grouped, 22 takeaway items not representative of all

		takeaway food consumption		compared to SES*; education, household income	educated counterparts, household income was not associated with overall takeaway consumption	takeaway foods, FFQ* prone to bias, Brisbane participants not representative of other populations
Reidpath et al. (2002) Cross-sectional	Australia	Examine the association between an area measure of SES* and the density of fast food outlets.	Populations from 267 postal districts obtained from the Australian Bureau of Statistics' 1996 Census data	Location of outlets identified by telephone directory, density of fast food outlets within an income category was combined population of all postal districts in an income category divided by total number of fast food franchises within those districts	Those living in the poorest areas were 2.5 times more exposed to fast food outlets than those in wealthier areas and those living in the very richest areas had no exposure to fast food outlets within their postal districts	Cross-sectional design precludes the ability to infer causal relationships, the greater density of fast food outlets in lower SES* areas cannot be a definite cause of the observed obesity, the existence of possible confounders, no individual data
Polsky et al. (2016) Cross-sectional	Canada	Absolute and relative densities of fast food versus other restaurants in relation to weight status	10,199 adults (aged 18+ years) residing in urban, residential areas of 4 cities in southern Ontario, Canada, from Canadian Community Health Survey	Data on restaurant locations were from a commercial database, fast food restaurant defined as locally owned or chain limited-service restaurant, restaurant density calculated using GIS*	Where fast food was the predominant type of restaurant, obesity figures for those living close to ≥5 fast food outlets was 2.5 times greater than the average	Cross-sectional design precludes the ability to infer causal relationships between restaurant exposure and weight, individuals with high BMI and who like fast food may self-select into neighbourhoods that have high exposure to fast food
Schneider and Gruber (2012) Cross-sectional	Germany	Neighbourhood deprivation and outlet density for tobacco, alcohol and fast food	92,000 inhabitants from 18 social areas in four districts in Cologne, Germany	Total number of fast food outlets recorded and visualized using GIS*, area affluence measured by the percentage of parents with children of nursery or school age with joint annual taxable income < 12,272 euros	The lower the income district the significantly higher availability of health damaging sources including fast food (P 0.009), tobacco (P 0.012) and alcohol (P 0.049); this correlation was strongest for fast food	Cross-sectional design precludes the ability to infer causal relationships, individuals may self-select to live in areas where they have access to their preferred products, absence of data on intake or frequency of fast food consumption
Pearce et al. (2007)	New Zealand	Neighbourhood Deprivation and	38,350 meshblocks across New	Data were geocoded and GIS* used to calculate	Access to fast food outlets in New Zealand was	Cross-sectional design precludes the ability to

Cross-sectional		Access to Fast Food Retailing	Zealand, each representing approximately 100 people	travel distances from each census meshblock (i.e., neighbourhood), and each school, to the closest fast food outlet	significantly higher (P < 0.001) in more deprived neighbourhoods, distance was at least 2 times further for the least deprived compared to the most deprived areas	infer causal relationships, lack of data on individual dietary intake or anthropometrics
Barton et al. (2015) Longitudinal	UK	Explore the association between diet and SES* and investigate trends in SES* inequalities in Scottish diet	11,374 individuals from 5020 Scottish households (over the period 2001–2009)	UK food purchase data to estimate household-level consumption data, detailed 14 day diary of all foods and beverages purchased for consumption both in and out of the home	Consumption of takeaway foods was significantly higher (P 0·008) in the most deprived quintile (24.2 g/person per d in the most compared to 18.3 g/person per d in the least) of the Scottish Index of Multiple Deprivation	Lower mean consumption for 'healthier foods' (e.g. wholemeal bread and oily fish) in more deprived quintiles was due to large numbers of non-consumers than was found in less deprived quintiles
Pieroni and Salmasi (2014) Cross-sectional	UK	Examine the role of fast food consumption on body weight	13,230 individuals each year (2004 and 2006) from UK wide involved in the British Household Panel Survey	Quantile regression approach to estimate correlations of body weight with several socio-economic determinants	Individuals with higher BMI, especially women, were more likely to live in areas with increased fast food exposure, relative prices of takeaway meals were correlated with obese/overweight adults	Cross-sectional design precludes the ability to infer causal relationships, a number of confounding variables were present
Athens et al. (2016) Cross-sectional	US	Test the independent relationship of access to fast food outlets and full-service supermarkets on frequency of fast food consumption	3,335 adults in Philadelphia and Baltimore in 2009 and 2010	Random-digit-dial telephone survey and point-of-purchase survey to compare respondent results, weekly fast food dining frequency recoded, residential intersections were geocoded and location of fast food outlets were calculated	Supermarket access was associated with declined fast food outlet use in telephone survey participants. > 1 fast food outlet within a 1-mile buffer was an independent predictor of increased fast food consumption among point-of-purchase participants	Cross-sectional design precludes the ability to infer causal relationships, level of attrition and location misclassification during geocoding may have caused bias in the data
Moore et al. (2009) Cross-sectional	US	Examine links between fast food consumption	5,633 men and women (aged 45–84 years) from 6	Self-administered 120-item FFQ*, diet quality measured against Alternate Healthy Eating	Non-fast food consumers were 2-3 times more likely to have a healthy diet than those eating fast food > 1	Self-reported data susceptible to recall bias, FFQ* may not capture diet variability, causation

		and diet, neighbourhood exposure to fast foods and consumption near home	study sites across the US	Index and the Western-type dietary pattern, fast food neighbourhood density using GIS*	times/week, greater fast food exposure; consumption of fast food near home increased 11%–61% and healthy diet decreased 3%–17%	cannot be inferred from these results, no data on types and quality of foods eaten at fast food outlets

***Abbreviations*: SES, Socio-economic status, FFQ, food frequency questionnaire; GIS, geographic information system.**

9.3 TABLE 3. SUMMARY OF STUDIES INVESTIGATING TAKEAWAY FOOD CONSUMPTION AND DEMOGRAPHIC INFLUENCES

Authors, type of study	Country	Study aim	Participants and setting	Methods	Results	Limitations
Smith et al. (2009) Cross-sectional	Australia	Examine takeaway food consumption in a national study of young Australian adults	2,862 (1,277 men, 1,585 women) aged 26–36 years involved in The Childhood Determinants of Adult Health Study	127-item FFQ* to record habitual food intake and takeaway food consumption, dietary intake compared to recommendations in the Australian Guide to Healthy Eating, anthropometrics measured	More men (37.9%) consumed takeaway food frequently than women (17.7%, $P < 0.001$), takeaway food consumption was associated with moderate weight, women who consumed takeaway > 2 had higher prevalence of overweight/obesity	Level of attrition was high, therefore, sample bias may have occurred, underestimated and overestimated dietary intake results from FFQ*
Mercille et al. (2016) Cross-sectional	Canada	Examine associations between exposure to neighbourhood food sources/consumption and diet knowledge of	722 women and men living in Montreal and Laval islands (2003–2005)	78-item FFQ* to record usual diet, PCA* to derive dietary patterns, person-level data on dietary knowledge from the Longitudinal Study on NuAge* cohort, GIS* for location mapping	Lower healthy diet scores associated with increased exposure to fast food outlets in men (β −0.18, P 0.02), the same relationship was present among women with low dietary knowledge (β −0.22, $P < 0.01$)	Cross-sectional design precludes the ability to infer causal relationships, older adults in NuAge* cohort not representative of other adults in Canada, FFQ dietary assessment prone to bias

		older women and men				
Srivastava (2015) Cross-sectional	India	Examine demographics of Indian consumer perception with respect to fast food chain restaurants	542 (379 males, 163 females) aged 12+ years consisting of baby boomers, X generation, and Y generation, living in Mumbai	Theory of the trust-commitment-loyalty explanation chain, ten-point semantic differential scales on quality, price, food etc., systematic sampling, demographic profiles recorded	Women, individuals with higher income, age group 19-25 and west Indians were more likely to visit global fast food chains than Indian chains, loyalty towards global brands due to a sense of increased quality	Small sample size, only India studied, therefore results may not be representative of other nations
Adams et al. (2015) Longitudinal	UK	Who eats out-of-home food; frequency and socio-demographic correlates of eating meals out and take-away meals at home	2,083 adults and 2,073 children from the UK National Diet and Nutrition Survey waves 1–4 (2008–12)	Interview with researcher on socio-demographics and shopping, cooking and eating habits; 4 day food diary; and nurse visit, parents or care providers gave information on children aged < 11 years	Eating meals out was associated with being in the 19–29 years age group, over 20% of children and adults ate takeaway meals at home once per week or more, girls were less likely to consume takeaway than boys	Data from UK may not be generalizable to others populations, no information collected on the specific type of meals out or take-away outlets visited
Hartmann et al. (2013) Cross-sectional and longitudinal	Switzerland	Develop cooking skill scale and examine relationship between cooking skill and consumption of various food groups	4,436 (47.2% male) participants from the Swiss Food Panel (2010 and 2011)	FFQ* to estimate habitual intake of various foods, cooking skills and psychological variables evaluated on a six-point scale	71% of women and 29% of men were responsible for meal cooking during the week, females had greater cooking skills than males, in men increased cooking skills correlated with cooking enjoyment rather than a responsibility	Cooking skills somewhat subjective as definition did not distinguish cooking from raw ingredients to cooking pre-prepared foods, results from FFQ* may be biased due to under/over-reporting
Fraser et al. (2012) Cross-sectional	UK	Analyse association between food outlet location, deprivation, weight status and ethnicity	1198 pregnant women from the BiB* Study	BiB* dataset included age, ethnicity, height and weight etc., food outlet details obtained from Bradford district council and business telephone directory, physical ‘groundthruthing’ to validate data, locations of	Over 95% of all participants lived within 500 m of a fast food outlet, individuals in higher deprived areas had greater access to fast food outlets and other food shops, fast food access (within 250m of residence) was	Cross-sectional design precludes the ability to infer causal relationships, large amounts of missing data especially in South Asian group, small sample in non-South Asian group

				fast food outlets mapped using GIS*	inversely associated with BMI in South Asians	
Dunn et al. (2012) Cross-sectional	US	Examine the effects of fast food availability on consumption and obesity among non-white and whites	1,000 (169 non-whites (self-reported black and Hispanic) and 831 whites) in Texas, US.	Data from the Brazos Valley Health Community Health Assessment Survey, frequency of fast food consumption, socio-demographic information recorded, fast food outlets mapped using GIS*	Non-whites were associated with increased obesity rates (49.1% versus 31.4% P < 0.01), greater access to (0.5 more outlets in 1 mile P < 0.01 and 3.3 more outlets in 3 miles P < 0.01) and consumption of fast food, increased availability of fast foods for whites was not associated with increased consumption or obesity risk	Cross-sectional design precludes the ability to infer causal relationships, proportion of whites to non-whites not equal, no data on dietary intake to show what participants were consuming
Lachat et al. (2009) Cross-sectional	Vietnam	Determine the nutritional contribution from 'out-of-home' foods and socio-economic factors in adolescents	1,172 adolescents living in urban and rural areas of Vietnam	24-hr dietary recall to record food intake, anthropometrics measured, SES* identified from a subsample of the population	'Out-of-home' foods contributed to 42% increased consumption of fruit and vegetables, increased consumption of sugar products (P < 0.001), was positively associated with having pocket money (P < 0.001)	Schools were not randomly selected which may limit external validity, Vietnamese adolescents may not represent adolescents from other nations

***Abbreviations*: BiB, Born in Bradford; FFQ, food frequency questionnaire; PCA, Principal component Analysis; NuAge, Nutrition and Successful Aging; GIS, geographic information system; SES, socio-economic status.**

9.4 TABLE 4. SUMMARY OF STUDIES INVESTIGATING DIETARY PATTERNS

Authors (Year) Study Title. Location	Participants (n =) and Inclusion criteria	Dietary Assessment Method	Dietary Pattern Analysis Method	Results	Limitations

Appannah et al. (2015) Identification of a dietary pattern associated with greater cardiometabolic risk in adolescence. **Perth, Australia.**	n = 1611 (87% participation) at 14 years of age n = 1009 (60% participation) at 17 years of age completed the food frequency questionnaire (FFQ), respectively. In the Western Australian Pregnancy (Raine) Study.	• Validated semi-quantitative FFQ was administered at 14 and 17 y of age to estimate habitual intake over previous year. • Selected frequency of consumption for each food was converted to a daily intake and linked with Australian Food Composition Tables to calculate average daily nutrient intakes.	• All food & drinks collapsed into 46 predefined food groups (47 groups at 17 years including alcohol) based on nutrient profiles or culinary usage. • Reduced rank regression data-dimension reduction technique that identifies DPs that are potentially relevant for the aetiology of disease • A-priori method to identify a nutrient-specific DP and the ability to control for dietary misreporting	Energy dense, high fat and low fibre DP is prospectively associated with unfavourable cardiometabolic risk factors	Estimated intakes of food groups and the response variables used in the DP analysis may have been affected by measurement and correlated errors inherent to the dietary assessment method. Residual confounding cannot be ruled out.
Batis et al. (2016) Using both principal component analysis and reduced rank regression to study dietary patterns and diabetes in Chinese adults. **China**	n = 4316 adults from the China Health and Nutrition Survey (CHNS)	• Combination of 24-hour dietary recall on 3 consecutive days with a household food inventory on the same 3 d. Start day was randomly allocated from Monday to Sunday. • Trained interviewers recorded the types and amounts of all food items consumed, the type of meal and the place of consumption. • For the food inventory, all foods in the household (purchased or produced at home) were measured on a daily basis with digital scales.	• PCA - Twenty-nine food groups. Most food groups had a high proportion of non-consumers possibly due to the fact that dietary intake was measured over a 3 d period. 18 food groups had < 30 % of consumers, 8 had 30–80 % of consumers and 3 had > 80 % of consumers. • Reduced rank regression	From the PCA; a modern high-wheat DP was positively associated with HbA1c. Traditional southern DP was negatively associated with HbA1c. Reduced rank regression pattern was closely related to the structure of both PCA DPs.	Only 3 d of intake. The proportion of consumers during the 3 d was very low (≤5 %) for many key food groups, such as processed meats, Western-style fast foods, salty snacks, ready-to-eat cereals and porridge, and calorically sweetened beverages.

Granic et al. (2015) Dietary Patterns and Socioeconomic Status in the Very Old: The Newcastle 85+ Study. **Newcastle, UK**	n = 793 (98.5% participation) in the Newcastle 85+ Study	• Two 24-hr multiple pass recall • Dietary intake was assessed on two different days of the week at least one week apart • No dietary recalls for Fridays and Saturdays	• Cluster analysis: Two-Step clustering and 30 food groups to derive DP • Multinomial logistic regression models to assess the association with socio-economic status.	Identified 3 distinct DP (characterised as 'High Red Meat', 'Low Meat', and 'High Butter') that varied with key socio-demographic, health and functioning measures.	Cross-sectional design and by a single measure of dietary exposure—therefore no causality can be inferred. Findings may not be generalizable to other populations of very old adults with dietary habits. Dietary misreporting
Mullie et al. (2010) Dietary patterns and socioeconomic position. **Belgium**	n = 1852 (37% participation) representative sample of military men from the Belgian Army (100 representative men took part in a pilot to test the validity of the FFQ (Mullie et al., 2009)).	• Mailed semi-quantitative FFQ with 150 food items. • Second self-reporting questionnaire used for health-related and lifestyle characteristics including smoking, marital status, main occupation, age, weight, height, number of children and knowledge of cardiovascular risk factors. • 1 of 9 frequency categories ranging from 'never' to 'more than 6 times per day'.	• Healthy Eating Index and Mediterranean Diet Score. The possible scores for Healthy Eating Index ranged from 0 to 100 and for Mediterranean Diet Score from 0 to 9, with a high score for the healthiest pattern. • PCA was applied to the data of the FFQ. First, 150 food items were classified into 34 predefined food groups with similar nutrient profile, according to Hu et al. (2000).	Higher income and education were associated with a higher score for Healthy Eating Index, Mediterranean Diet Score and Healthy DP. All three methods obtained a comparable ranking	Low response rate of 37% (responders were older than non-responders). PCA involves several arbitrary decisions. Representative for Belgian army men and could be generalisable for working Belgian men only
Barker et al. (1990) Dietary behaviours and sociocultural demographics in Northern Ireland. **Northern Ireland, UK**	n = 592 (258 men and 334 women) aged 16-64 years randomly selected from N.I. population	• 7 d weighed food record (all food and drink) • Social and personal data were collected by an interviewer-administered questionnaire. • Anthropometric data was obtained using appropriate equipment.	• Weights of a range of food groups were calculated and food data into 41 food groups • PCA on the correlation matrix for the 41 food groups. SPSS, routine FACTOR, used to identify the primary components which accounted for variation in dietary intake. • SPSS, routine ANOVA, was used to correlate DPs with	Generated 4 distinct DPs; 'traditional diet', 'cosmopolitan diet', 'convenience diet' and 'meat and two veg diet'	Dietary misreporting. Subjectivity in determining food groups before application of PCA. Findings may not be generalisable to other populations. No data on objective measures of food intake,

			sociocultural, lifestyle and anthropometric variables.		such as biomarkers of nutrient intake.
Funtikova et al. (2015) Modest validity and fair reproducibility of dietary patterns derived by cluster analysis. **Girona, Spain**	n = 107 (71.3% participation) of women and men aged 30 to 80 years completed the study. From a population-based cross-sectional survey in 2005	• FFQ at baseline (FFQ1) and after 1 year (FFQ2) • Reference method: Twelve 24-hour dietary recalls (24 h DR) monthly over a 12-month period by a trained telephone interviewer	• Cluster Analysis - K-mean cluster algorithm was used to derive DP from FFQ1, FFQ2, and 24 hDR.	Identified "fruits & vegetables" and "meat" DPs from the dietary data. κ statistic revealed a fair validity and reproducibility of clusters.	24 h DR & FFQ are retrospective and prone to similar limitations. Defined DP are population specific. No data on biomarkers of nutrient intake. Cluster analysis is prone to arbitrary decisions during the process of cluster definition. Small sample size makes it difficult to reach a high level of significance.
Ashby-Mitchell et al. (2015) Role of Dietary Pattern Analysis in Determining Cognitive Status in Elderly Australian Adults. **Australia**	n = 577 of the sample (age 60+ at baseline) that had completed FFQ and mini-mental state examination. From the Australian Diabetes, Obesity and Lifestyle Study (AusDiab), a population-based national survey of the general Australian population aged 25+ years	• AusDiab semi-quantitative FFQ consisted of 121 items (101 food items). • Assessed usual intake and recorded the amount and types of specific food items consumed by participants. • 1 of 10 frequency categories ranging from 'never' to 'three or more times per day'. • Average daily intake of food weight in grams was computed.	• Three methods were used in order to classify these foods before applying PCA. (1) The 101 individual food items were used (no categorisation). (2) and (3) The, foods were combined and reduced to 32 and 20 food groups, respectively, based on nutrient content and culinary usage—a method employed in several other published studies for PCA.	'Western' DP was predictive of poorer memory and processing speed. 'Vegetable, Grains and Wine' DP was a predictor of better processing speed. 'Prudent' DP was predictive of poorer processing speed	Dietary intake was self-reported. Subjectivity in determining food groups before application of PCA, but the method of food variable reduction employed has been widely used in other studies. Selection bias, as only older adults with dietary and cognitive data were included in the study, this limits the generalisability of the study's findings.

Reedy et al. (2010) Comparing 3 Dietary Pattern Methods—Cluster Analysis, Factor Analysis, and Index Analysis—With Colorectal Cancer Risk. **USA**	n = 492, 306 (293,576 men and 198,730 women) aged between 50 and 71 y and residents from 6 states (Florida, California, Louisiana, New Jersey, North Carolina, Pennsylvania) or 2 metropolitan areas (Atlanta, Georgia; Detroit, Michigan).	• 124-item FFQ, an early version of the Diet History Questionnaire, to assess dietary intake over the past year. • The Diet History Questionnaire was calibrated (and further validation was done with the AARP FFQ and two 24-hour recalls within the NIH–AARP Diet and Health Study	• 181 food groups based on the 204 food items drawn from the FFQ (because line items contain more than one food item, the final number of food items from the 124-item FFQ was 204). • Cluster analysis • Factor analysis • Index analysis	Cluster analysis & factor analysis accounted for the variation in intakes and how well those variances related to risk, whereas index analysis asked whether variation from a predefined diet related to risk. However, similarities were seen across all methods.	Dietary intake was self-reported and retrospective. No reference biomarkers for nutrients. FFQ prone to bias.
del mar Bibiloni (2012) Western and Mediterranean dietary patterns among Balearic Islands' adolescents: socio-economic and lifestyle determinants. **Balaeric Islands, Spain**	n = 1231 individuals (82% participation) of a random sample of the adolescent population 12–17 years.	• Dietary assessment was based on a 145-item semi-quantitative FFQ and two non-consecutive 24 h DR (one in the warm season (May– September) and one in the cold season (November–March) to account for the influence of seasonal variations) • Anthropometric measurements and questions related to socio-economic, lifestyle, physical activity and body image were assessed.	• PCA and varimax rotation were run on Z-scored transformed food consumption variables in order to identify salient food patterns. • Only food groups with factor loading > 0.250 were retained for each factor. • Multiple logistic regression analyses were used to simultaneously examine the effect of different socio-economic and lifestyle variables on the DPs.	PCA identified two major DPs among adolescents ('Western' and 'Mediterranean'). Low parental socio-economic status, much leisure-time on sedentary behaviours such as media screen time and body image are factors associated with the 'Western' DP.	Dietary and physical activity data were self-reported. Dietary under-reporting is usually associated with gender and weight status. Physical activity over-reporting due to social desirability bias. The FFQ did not differentiate between wholegrain and white bread. Factor analysis is data-specific, therefore, findings may not be generalisable to other populations.

9.5 Ethical approval for PhD project (Approved 11th January 2016)

Reply Reply All Forward

Mon 11/01/2016 12:38

Williams, Mandy

Ethical Approval

To Janssen, Hayley

Cc Stevenson, Leo; Davies, Ian

Follow up. Start by 11 January 2016. Due by 11 January 2016.
You replied to this message on 11/01/2016 13:54.
This message was sent with High importance.

Dear Hayley

With reference to your application for Ethical Approval

16/EHC/002 - Hayley Janssen (PGR) - Determinants of Takeaway Food Consumption in Relation to Dietary Patterns (Leo Stevenson/Ian Davies)

Liverpool John Moores University Research Ethics Committee (REC) has considered the above application and I am pleased to inform you that ethical approval has been granted and the study can now commence.

Approval is given on the understanding that:

- any adverse reactions/events which take place during the course of the project are reported to the Committee immediately;
- any unforeseen ethical issues arising during the course of the project will be reported to the Committee immediately;
- the LJMU logo is used for all documentation relating to participant recruitment and participation e.g. poster, information sheets, consent forms, questionnaires. The LJMU logo can be accessed at http://www.ljmu.ac.uk/corporatecommunications/60486.htm

Where any substantive amendments are proposed to the protocol or study procedures further ethical approval must be sought.

Applicants should note that where relevant appropriate gatekeeper / management permission must be obtained prior to the study commencing at the study site concerned.

For details on how to report adverse events or request ethical approval of major amendments please refer to the information provided at http://www.ljmu.ac.uk/RGSO/93205.htm

Please note that ethical approval is given for a period of five years from the date granted and therefore the expiry date for this project will be January 2021. An application for extension of approval must be submitted if the project continues after this date.

Yours sincerely

LIVERPOOL JOHN MOORES UNIVERSITY

Mandy Williams, Research Support Officer
(Research Ethics and Governance)
Research and Innovation Services
Kingsway House, Hatton Garden, Liverpool L3 2AJ
t: 01519046467 e: a.f.williams@ljmu.ac.uk

9.6 ETHICS MAJOR AMENDMENT 1 - ABERYSTWYTH COLLABORATION FOR METABOLOMICS RESEARCH, UPDATE OF STUDY MATERIALS AND RECRUITMENT METHODS (APPROVED 10TH MARCH 2016)

Reply Reply All Forward

Thu 10/03/2016 15:59

Williams, Mandy

Major Amendment Approval - 16/EHC/002

To Janssen, Hayley

Follow up. Start by 11 March 2016. Due by 11 March 2016.
You forwarded this message on 02/06/2016 08:38.

Dear Hayley

Further to the above applications for major amendments which you recently submitted for consideration by the University's Research Ethics Committee. Please accept this email as formal confirmation that REC agreed to approve this application by Chairs action.

LIVERPOOL JOHN MOORES UNIVERSITY

Mandy Williams, Research Support Officer
(Research Ethics and Governance)
Research and Innovation Services
Kingsway House, Hatton Garden, Liverpool L3 2AJ
t: 01519046467 e: a.f.williams@ljmu.ac.uk

9.7 Ethics major amendment 2 – Recruitment technqiues (Approved 4th October 2016

Tue 04/10/2016 14:35

Harriss, Dave

FW: Major amendment

To Janssen, Hayley

Cc Stevenson, Leo; Williams, Mandy; **researchethics**

Follow up. Start by 05 October 2016. Due by 05 October 2016.
You replied to this message on 04/10/2016 14:36.

Dear Hayley

Please accept this email as approval notification of the study amendments.

Thank you for notifying UREC of the changes – it is much appreciated.

Best wishes
Dave

Dr Dave Harriss
Research Governance Manager
Senior Lecturer in Sport & Exercise Sciences
Liverpool John Moores University
07929999021 (work mobile number)

From: Janssen, Hayley
Sent: 04 October 2016 14:28
To: Harriss, Dave <D.Harriss@ljmu.ac.uk>
Cc: Stevenson, Leo <L.Stevenson@ljmu.ac.uk>; Williams, Mandy <A.F.Williams@ljmu.ac.uk>
Subject: Major amendment

Hi Dave,

Please could you review this major amendment (see attached form) for my PhD study (16/EHC/002). I have also highlighted all changes in yellow on the supporting materials.

Many thanks,
Hayley

9.8 ETHICS MAJOR AMENDMENT 3 – PERSONAL INFORMATION AND STUDY FLYER (APPROVED 10TH NOVEMBER 2016)

Reply Reply All Forward

Thu 10/11/2016 14:57

Harriss, Dave

FW: Notification of Amendment (16/EHC/002)

To Janssen, Hayley

Cc Williams, Mandy; **researchethics**

Follow up. Start by 13 June 2018. Due by 13 June 2018.
You replied to this message on 10/11/2016 15:02.

Dear Hayley

Thank you for notifying UREC about the study amendments – much appreciated. Please accept this email as notification of ethical approval.

Mandy – can you please add this to the records.

Best wishes
Dave

Dr Dave Harriss
Research Governance Manager
Chair of the LJMU Research Ethics Committee
Senior Lecturer in Sport & Exercise Sciences
Liverpool John Moores University
07929999021 (work mobile number)
0151 9046236 (office, TRB 1.17)

From: Janssen, Hayley
Sent: 10 November 2016 12:16
To: researchethics
Cc: Williams, Mandy; Richardson, Lucy; Davies, Ian
Subject: Notification of Amendment (16/EHC/002)

Dear Research Ethics Committee,

I am submitting a notification of amendment to my Approved Ethics Application (16/EHC/002).

Attached is a single pdf file containing my notification of amendment and all supporting documentation. All changes to the supporting documents have been highlighted in yellow. I have also included a copy of my supervisory conformation letter to support my notification of amendment.

Kind regards,

9.9 Ethics major amendment 4 – Changes to Study 3 materials (Approved 7th February 2017)

Reply | Reply All | Forward

Tue 07/02/2017 14:24

Williams, Mandy

Major Amendment Approval - 16/EHC/002

To Janssen, Hayley

Dear Hayley

Further to the above applications for major amendments which you recently submitted for consideration by the University's Research Ethics Committee. Please accept this email as formal confirmation that REC agreed to approve this application by Chairs action.

LIVERPOOL JOHN MOORES UNIVERSITY

Mandy Williams, Research Support Officer
(Research Ethics and Governance)
Research and Innovation Services
Kingsway House, Hatton Garden, Liverpool L3 2AJ
t: 01519046467 e: a.f.williams@ljmu.ac.uk

9.10 Ethics major amendment 5 – Acceptability questionnaire (Approved 9th November 2017)

Reply | Reply All | Forward

Thu 09/11/2017 17:21

Williams, Mandy

Major Amendment Approval - 16/EHC/002

To Janssen, Hayley

Cc **researchethics**

You replied to this message on 09/11/2017 17:37.

Dear Hayley

Further to the above applications for major amendments which you recently submitted for consideration by the University's Research Ethics Committee. Please accept this email as formal confirmation that REC agreed to approve this application by Chairs action.

LIVERPOOL JOHN MOORES UNIVERSITY

Mandy Williams, Research Support Officer
(Research Ethics and Governance)
Research and Innovation Services
Kingsway House, Hatton Garden, Liverpool L3 2AJ
t: 01519046467 e: a.f.williams@ljmu.ac.uk

9.11 Study 1 Recruitment letter

Liverpool John Moores University
IM Marsh Campus
Barkhill Road
Aigburth
Liverpool
L17 6BD

Dear Participant,

You are invited to take part in a research study by Liverpool John Moores University entitled '**Determinants of Takeaway Food Consumption**'

A participant information sheet and informed consent form have been enclosed with this letter for you to read through outlining the study in more detail. If you decide you would like to take part in the study it would be greatly appreciated if you could return the completed informed consent form and personal details form in the envelope provided. If you choose to participate in the study you will be contacted in due course.

If you would like to contact a member of the research team to discuss the study further, please use the details below:-

Hayley Janssen (Postgraduate Research Student)
Liverpool John Moores University
IM Marsh Campus
Barkhill Road
Aigburth
Liverpool
L17 6BD
(Email – H.G.Janssen@2012.ljmu.ac.uk)

Leo Stevenson (Senior Lecturer)
Liverpool John Moores University
IM Marsh Campus
Barkhill Road
Aigburth
Liverpool
L17 6BD
(Email – L.Stevenson@ljmu.ac.uk)

Thank you very much for your help with this research.

Kind regards,
Hayley Janssen

9.12 Study 1 Participant information sheet (hardcopy)

Participant Information Sheet

Study Title: Determinants of Takeaway Food Consumption

By Hayley Janssen (Faculty of Education, Health and Community, LJMU)

You are invited to take part in a research project. Before you decide, it is important that you understand the purpose of the research and what it involves. Therefore, please take time to read the following information and decide if you want to take part or not. If there is anything unclear or you would like to obtain more information, please do not hesitate to ask us.

1. What is the purpose of the study?

This study will investigate patterns of takeaway food consumption among people in Liverpool. It will also investigate factors that may influence takeaway food consumption.

2. Do I have to take part?

No. It is up to you to decide whether or not to take part. Participation in this study is voluntary. If you decide to participate you are still free to withdraw at any time without giving a reason.

3. What will happen to me if I take part?

If you agree to take part you will be:

- Asked to provide some personal details (e.g. name and contact details) to take part in both phases of the study.
- Asked to complete a food frequency questionnaire in person, via the post or online, which will give an insight into your dietary/takeaway food intake, demographical characteristics (e.g. age, gender and occupation) and self-reported body measurements (e.g. height and weight). This questionnaire will take approximately 20-30 minutes to complete.
- Contacted by telephone to take part in three recorded interviews regarding your takeaway food consumption. These interviews will take approximately 10-20 minutes each time.
- May be contacted after the dietary survey to take part in the second part of the study which includes a face to face interview with the researcher. The face to face interview may take up to an hour but the interviewer is happy to spend less or more time – it is up to you.

- Once the study is complete you will be debriefed on your role in the study and given a chance to ask any questions.

Please be aware that you may be excluded from the study if you are under the age of 18 or over 65 and if you do not live in Liverpool - you should make the researcher aware of this before beginning the study!

4. Are there any risks/benefits involved?

You will be entered into a prize draw to win an iPad after completing the questionnaire and again to another prize draw to win an iPad at the end of the study (if you take part). You will be asked to fill in the questionnaire honestly, all information obtained as part of the study will be made anonymous so the answers provided by you will be unidentifiable in any further publications. The wider benefits are that this preliminary evidence may help to assist the development of interventions and public health strategies to reduce the consumption of takeaway foods or improve the nutritional composition of takeaway food.

5. Will my taking part in the study be kept confidential?

Completion of the study is voluntary and anonymous; the information provided by you will be unidentifiable in any publications. Any personal information collected during this study will be stored securely on password protected computers or in locked filing cabinets.

6. Contact Details of Researcher

If you have any questions regarding your participation or the study please do not hesitate to contact:

Hayley Janssen, e-mail: **H.G.Janssen@2012.ljmu.ac.uk**
Leo Stevenson, e-mail: **L.Stevenson@ljmu.ac.uk**

If you any concerns regarding your involvement in this research, please discuss these with the researcher in the first instance. If you wish to make a complaint, please contact researchethics@ljmu.ac.uk and your communication will be re-directed to an independent person as appropriate.

Note: A copy of the participant information sheet should be retained by the participant with a copy of the signed consent form

Informed Consent Form

Study Title: Determinants of Takeaway Food Consumption

Researchers name - Hayley Janssen (Faculty of Education, Health and Community, LJMU)
Contact details: H.G.Janssen@2012.ljmu.ac.uk

1. I confirm that I have read and understand the information provided for the above study. I have had the opportunity to consider the information, ask questions and have had these answered satisfactorily ☐

2. I understand that my participation is voluntary and that I am free to withdraw at any time during data collection, without giving a reason and that this will not affect my legal rights ☐

3. I understand that any personal information collected during the study will be anonymised and remain confidential ☐

4. I understand that I will be invited to fill out a questionnaire regarding my dietary and takeaway food intake, some demographic and personal information and that I may be invited to take part in three 'Multiple Pass' 24-hour dietary recalls and an interview with the researcher. ☐

5. I consent to be contacted after completing the questionnaire to take part in an interview with the researcher to discuss reasons for consuming takeaway food. ☐

6. I agree to take part in the above study ☐

Name of Participant   Date   Signature

Name of Researcher   Date   Signature

Name of Person taking consent   Date   Signature
(if different from researcher)
Note: When completed 1 copy for participant and 1 copy for researcher

For office use only. Participant number: ………….

9.14 Study 1 Amended Version of the EPIC-Norfolk printed semi-quantitative FFQ with self-reported takeaway food consumption, demographic, lifestyle and anthropometric questions (used as hardcopy and online on Online Surveys)

LIVERPOOL JOHN MOORES UNIVERSITY

Food Frequency Questionnaire

Name:_____________________________

Date:______________________________

Thank you for agreeing to take part in this study.

This is a simple questionnaire designed to help us understand what foods you normally eat. It is not a test, so there are no right or wrong answers. It is your usual diet we are interested in.

All information will be treated in the strictest confidence.

Please turn over and read the instructions for answering questions before completing the questionnaire. Please answer every question. If you are uncertain about how to answer a question then do the best you can. Thank you for your time.

ID							
	Date	d	d	m	m	y	y

For office use only

FFQ Questionnaire

About the food you eat

The following questions are about the food you usually eat and how often you eat certain foods. Please read the following instructions before answering the questions. For each food there is an amount shown, either a "medium serving" or a common household unit such as a slice or teaspoon. Please put a tick (✓) in the box to indicate how often, **on average**, you have eaten the specified amount of each food **during the past year.**

EXAMPLE:

For white bread the amount is one slice, so if you ate 4 or 5 slices a day, you should put a tick in the column headed "4-5 per day".

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
BREAD & SAVOURY BISCUITS (one slice or biscuit)	Never or less than once/month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
White bread and rolls								✓	

EXAMPLE:

For chips, the amount is a "medium serving", so if you had a helping of chips twice a week you should put a tick in the column headed "2-4 per week".

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
POTATOES, RICE & PASTA (medium serving)	Never or less than once/month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Chips				✓					

It is now your turn to answer ☺

Please put a tick (✓) in each box to indicate how often, **on average**, you have eaten each food **during the past year.**

Please estimate your average food use as best you can, and please answer every question - do not leave **ANY** lines blank. ***PLEASE PUT A TICK (✓) ON EVERY LINE.***

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
1. MEAT & FISH ***(medium serving)*** ***(not fast food or takeaway)***	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Beef: roast, steak, mince, stew casserole, curry or bolognese	1	2	3	4	5	6	7	8	9
Beef burgers (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
Pork: roast, chops, stew, slice or curry	1	2	3	4	5	6	7	8	9
Lamb: roast, chops, stew or curry	1	2	3	4	5	6	7	8	9
Chicken, turkey or other poultry: including fried, casseroles or curry	1	2	3	4	5	6	7	8	9
Bacon	1	2	3	4	5	6	7	8	9
Ham	1	2	3	4	5	6	7	8	9
Corned beef, Spam, luncheon meats	1	2	3	4	5	6	7	8	9
Sausages	1	2	3	4	5	6	7	8	9
Savoury pies, e.g. meat pie, pork pie, pasties, steak & kidney pie, sausage rolls, scotch egg	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	**AVERAGE USE LAST YEAR**								
1. MEAT & FISH,(continued) ***(medium serving)*** ***(not fast food or takeaway)***	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Liver, liver pate, liver sausage	1	2	3	4	5	6	7	8	9
Fried fish in batter, as in fish and chips (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
Fish fingers, fish cakes	1	2	3	4	5	6	7	8	9
Other white fish, fresh or frozen, e.g. cod, haddock, plaice, sole, halibut	1	2	3	4	5	6	7	8	9
Oily fish, fresh or canned, e.g. mackerel, kippers, tuna, salmon, sardines, herring	1	2	3	4	5	6	7	8	9
Shellfish, e.g. crab, prawns, mussels	1	2	3	4	5	6	7	8	9
2. BREAD & SAVOURY BISCUITS ***(one slice or biscuit)*** ***(not fast food or takeaway)***									
White bread and rolls	1	2	3	4	5	6	7	8	9
Scones, teacakes, crumpets, muffins or croissants	1	2	3	4	5	6	7	8	9
Brown bread and rolls	1	2	3	4	5	6	7	8	9
Wholemeal bread and rolls	1	2	3	4	5	6	7	8	9
Cream crackers, cheese biscuits	1	2	3	4	5	6	7	8	9
Pitta bread, naan bread, chapatti (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
Garlic bread (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
3. CEREALS ***(one bowl)***	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Porridge, Readybrek	1	2	3	4	5	6	7	8	9
Sugar coated cereals e.g. Sugar Puffs, Cocoa Pops, Frosties	1	2	3	4	5	6	7	8	9
Non-sugar coated cereals e.g. Cornflakes, Rice Crispies	1	2	3	4	5	6	7	8	9
All Bran, Bran Flakes, Muesli	1	2	3	4	5	6	7	8	9
Wholegrain cereals e.g. Cheerios, Weetabix, Shredded Wheat	1	2	3	4	5	6	7	8	9
4. POTATOES, RICE & PASTA ***(medium serving)*** ***(not fast food or takeaway)***									
Boiled, mashed, instant or jacket potatoes	1	2	3	4	5	6	7	8	9
Chips, potato waffles, potato wedges (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
Roast potatoes	1	2	3	4	5	6	7	8	9
Yorkshire pudding, pancakes, dumpling	1	2	3	4	5	6	7	8	9
Potato salad	1	2	3	4	5	6	7	8	9
White rice	1	2	3	4	5	6	7	8	9
Brown rice	1	2	3	4	5	6	7	8	9
White or green pasta, e.g. spaghetti, macaroni, noodles	1	2	3	4	5	6	7	8	9
Tinned pasta, e.g. spaghetti, ravioli, macaroni	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
4. POTATOES, RICE & PASTA (continued) ***(medium serving)*** ***(not fast food or takeaway)***	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Super noodles, pot noodles, pot savouries	1	2	3	4	5	6	7	8	9
Wholemeal pasta	1	2	3	4	5	6	7	8	9
Lasagne, moussaka, cannelloni	1	2	3	4	5	6	7	8	9
Pizza (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
5. DAIRY PRODUCTS & FATS									
Single or sour cream *(tablespoon)*	1	2	3	4	5	6	7	8	9
Double or clotted cream *(tablespoon)*	1	2	3	4	5	6	7	8	9
Low fat yoghurt, low fat greek yoghurt, fromage frais *(125g carton)*	1	2	3	4	5	6	7	8	9
Full fat or Greek yoghurt *(125g carton)*	1	2	3	4	5	6	7	8	9
Dairy desserts *(125g carton),* e.g. mousse or custard	1	2	3	4	5	6	7	8	9
Cheese, e.g. Cheddar, Brie, Edam *(medium serving)*	1	2	3	4	5	6	7	8	9
Cottage cheese, low fat soft cheese *(medium serving)*	1	2	3	4	5	6	7	8	9
Eggs as boiled, fried, scrambled, poached, omelette etc. *(one)*	1	2	3	4	5	6	7	8	9
Quiche *(medium serving)*	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	**AVERAGE USE LAST YEAR**								
5.(b) The following on bread or vegetables *(teaspoon)*	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Butter	1	2	3	4	5	6	7	8	9
Block margarine, e.g. Stork, Krona	1	2	3	4	5	6	7	8	9
Polyunsaturated margarine, e.g. Flora sunflower	1	2	3	4	5	6	7	8	9
Other soft margarine, dairy spreads, e.g. Blue Band, Clover	1	2	3	4	5	6	7	8	9
Low fat spread, e.g. Gold	1	2	3	4	5	6	7	8	9
6. SWEETS & SNACKS									
Sweet biscuits, chocolate, e.g. digestive *(one)*	1	2	3	4	5	6	7	8	9
Sweet biscuits, plain, e.g. Nice, ginger *(one)*	1	2	3	4	5	6	7	8	9
Cakes e.g. fruit, sponge, sponge pudding *(medium serving)*	1	2	3	4	5	6	7	8	9
Sweet buns & pastries e.g. flapjacks, doughnuts, Danish pastries, cream cakes *(medium serving)*	1	2	3	4	5	6	7	8	9
Fruit pies, tarts, crumbles *(medium serving)*	1	2	3	4	5	6	7	8	9
Milk puddings, e.g. rice, custard, trifle *(medium serving)*	1	2	3	4	5	6	7	8	9
Ice cream, choc ices *(one)*	1	2	3	4	5	6	7	8	9
Chocolates *(small bar or ¼ pound of chocolates)*	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	**AVERAGE USE LAST YEAR**								
6. SWEETS & SNACKS (continued)	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Sweets, toffees, mints *(one packet)*	1	2	3	4	5	6	7	8	9
Sugar added to tea, coffee, cereal *(teaspoon)*	1	2	3	4	5	6	7	8	9
Crisps or other packet snacks e.g. Wotsits *(one packet)*	1	2	3	4	5	6	7	8	9
Peanuts or other nuts *(one packet)*	1	2	3	4	5	6	7	8	9
7. SOUPS, SAUCES AND SPREADS									
Vegetable soups *(bowl)*	1	2	3	4	5	6	7	8	9
Meat soups *(bowl)*	1	2	3	4	5	6	7	8	9
Sauces, e.g. white sauce, cheese sauce, gravy *(medium serving)*	1	2	3	4	5	6	7	8	9
Tomato based sauces e.g. pasta sauces *(medium serving)*	1	2	3	4	5	6	7	8	9
Tomato ketchup, brown sauce *(tablespoon)*	1	2	3	4	5	6	7	8	9
Relishes e. g. pickles, chutney, mustard *(tablespoon)*	1	2	3	4	5	6	7	8	9
Low calorie, low fat salad cream or mayonnaise *(tablespoon)*	1	2	3	4	5	6	7	8	9
Salad cream, mayonnaise *(tablespoon)*	1	2	3	4	5	6	7	8	9
French dressing *(tablespoon)*	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	**AVERAGE USE LAST YEAR**								
7. SOUPS, SAUCES AND SPREADS (continued)	Never or less than once/ month	1-3 per month	Once A Week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Other salad dressing *(tablespoon)*	1	2	3	4	5	6	7	8	9
Marmite, Bovril *(teaspoon)*	1	2	3	4	5	6	7	8	9
Jam, marmalade, honey, syrup *(teaspoon)*	1	2	3	4	5	6	7	8	9
Peanut butter *(teaspoon)*	1	2	3	4	5	6	7	8	9
Chocolate spread, chocolate nut spread e.g. nutella *(teaspoon)*	1	2	3	4	5	6	7	8	9
Dips e.g. hummus, cheese and chive *(tablespoon)*	1	2	3	4	5	6	7	8	9
8. DRINKS									
Tea *(cup)*	1	2	3	4	5	6	7	8	9
Coffee, instant or ground *(cup)*	1	2	3	4	5	6	7	8	9
Coffee whitener, e.g. Coffee-mate *(teaspoon)*	1	2	3	4	5	6	7	8	9
Cocoa, hot chocolate *(cup)*	1	2	3	4	5	6	7	8	9
Horlicks, Ovaltine *(cup)*	1	2	3	4	5	6	7	8	9
Wine *(glass)*	1	2	3	4	5	6	7	8	9
Beer, lager or cider *(half pint)*	1	2	3	4	5	6	7	8	9
Port, sherry, vermouth, liqueurs *(glass)*	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
8. DRINKS (continued)	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Spirits, e.g. gin, brandy, whisky, vodka *(single)*	1	2	3	4	5	6	7	8	9
Low calorie or diet fizzy soft drinks *(glass)*	1	2	3	4	5	6	7	8	9
Fizzy soft drinks, e.g. Coca cola, lemonade *(glass)*	1	2	3	4	5	6	7	8	9
Pure fruit juice (100%) e.g. orange, apple juice *(glass)*	1	2	3	4	5	6	7	8	9
Fruit squash or cordial *(glass)*	1	2	3	4	5	6	7	8	9
9. FRUIT *(1 fruit or medium serving)* *For very seasonal fruits such as strawberries, please estimate your average use when the fruit is in season									
Apples	1	2	3	4	5	6	7	8	9
Pears	1	2	3	4	5	6	7	8	9
Oranges, satsumas, mandarins, tangerines, clementines	1	2	3	4	5	6	7	8	9
Grapefruit	1	2	3	4	5	6	7	8	9
Bananas	1	2	3	4	5	6	7	8	9
Grapes	1	2	3	4	5	6	7	8	9
Melon	1	2	3	4	5	6	7	8	9
*Peaches, plums, apricots, nectarines	1	2	3	4	5	6	7	8	9
*Strawberries, raspberries, kiwi fruit	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
9. FRUIT (continued) ***(1 fruit or medium serving)***	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Tinned fruit	1	2	3	4	5	6	7	8	9
Dried fruit, e.g. raisins, prunes, figs	1	2	3	4	5	6	7	8	9
10. VEGETABLES Fresh, frozen or tinned ***(medium serving)***									
Carrots	1	2	3	4	5	6	7	8	9
Spinach	1	2	3	4	5	6	7	8	9
Broccoli	1	2	3	4	5	6	7	8	9
Brussels sprouts	1	2	3	4	5	6	7	8	9
Cabbage	1	2	3	4	5	6	7	8	9
Peas	1	2	3	4	5	6	7	8	9
Green beans, broad beans, runner beans	1	2	3	4	5	6	7	8	9
Marrow, courgettes	1	2	3	4	5	6	7	8	9
Cauliflower	1	2	3	4	5	6	7	8	9
Parsnips, turnips, swedes	1	2	3	4	5	6	7	8	9
Leeks	1	2	3	4	5	6	7	8	9
Onions	1	2	3	4	5	6	7	8	9
Garlic	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
10. VEGETABLES Fresh, frozen or tinned (continued) ***(medium serving)***	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Mushrooms	1	2	3	4	5	6	7	8	9
Sweet peppers	1	2	3	4	5	6	7	8	9
Beansprouts	1	2	3	4	5	6	7	8	9
Green salad, lettuce, cucumber, celery	1	2	3	4	5	6	7	8	9
Mixed vegetables (frozen or tinned)	1	2	3	4	5	6	7	8	9
Watercress	1	2	3	4	5	6	7	8	9
Tomatoes	1	2	3	4	5	6	7	8	9
Sweetcorn	1	2	3	4	5	6	7	8	9
Beetroot, radishes	1	2	3	4	5	6	7	8	9
Coleslaw	1	2	3	4	5	6	7	8	9
Avocado	1	2	3	4	5	6	7	8	9
Baked Beans	1	2	3	4	5	6	7	8	9
Dried lentils, beans, peas	1	2	3	4	5	6	7	8	9
Tofu, soya meat, TVP, Vegeburger	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

YOUR DIET LAST YEAR, continued

11. (a) What type of milk did you most often use? *Select one answer*

Full cream……❒1
Channel Islands……❒2
Dried milk……❒3
Semi-skimmed……❒4
Skimmed……❒5
Soya……❒6
Other……❒7
None……❒8

12. (b) Approximately, how much milk did you drink each day, including milk with tea, coffee, cereals etc? *Select one answer*

None……❒1
Quarter of a pint (roughly 125mls)……❒2
Half a pint (roughly 250mls) ……❒3
Three quarters of a pint (roughly 375mls) ……❒4
One pint (roughly 500mls) ……❒5
More than one pint (more than 500mls) ……❒6

13. What kind of fat did you most often use for frying, roasting, grilling etc? *Select one answer*

Butter……❒1
Lard/dripping……❒2
Solid vegetable fat……❒3
Margarine……❒4
Vegetable oil……❒5
Olive oil……❒6
None……❒7

14. How often did you eat food that was fried at home? *Select one answer*

Daily……❒1
1-3 times a week……❒2
4-6 times a week……❒3
Less than once a week……❒4
Never……❒5

15. How often did you eat fried food *away* from home? *Select one answer*

Daily……❒1
1-3 times a week……❒2
4-6 times a week……❒3
Less than once a week……❒4
Never……❒5

16. **(a) How often did you add salt to food while cooking?**
Select one answer

Always……❒1
Usually……❒2
Sometimes……❒3
Rarely……❒4
Never……❒5

17. **(b) How often did you add salt to any food at the table?**
Select one answer

Always……❒1
Usually……❒2
Sometimes……❒3
Rarely……❒4
Never……❒5

18. **Do you follow a special diet?**
Please tick all that apply

No……❒1
Yes, because of a medical condition/allergy……❒2
Yes, to lose weight……❒3
Yes, because of personal beliefs (religion, vegetarian)……❒4
Yes, other……❒5

19. **Over the last year, how often have you eaten organic foods?**
Select one answer

Most days……❒1
Once or twice a week……❒2
Once a month……❒3
Never/hardly ever……❒4

20. **Have you taken any of the following during the past year?**

a) Vitamins (e.g. multivitamins, vitamin B, vitamin C, folic acid)

Yes……❒1
No……❒2

If YES, what type and when ____

b) Minerals (e.g. iron, calcium, zinc, magnesium)

Yes……❒1
No……❒2

If YES, what type and when ____

c) Fish oils (e.g. cod liver oil, omega-3)

Yes……❐1
No……❐2

If YES, what type and when ____

d) Other food supplements (e.g. oil of evening primose, starflower oil, royal jelly, ginseng)

Yes……❐1
No……❐2

If YES, what type and when ____

21. For the following foods, please list the top three makes and/or brands you most commonly consume.
If you do not eat this type of food please tick the 'not eaten' box.

Bread:

1:______________________________

2:______________________________

3:______________________________

Not eaten……❐

Breakfast Cereal:

1:______________________________

2:______________________________

3:______________________________

Not eaten……❐

About your takeaway food and/or fast food consumption

For the purpose of this survey, 'takeaway food' or 'fast food' can include foods purchased from small, individual outlets such as chip shops/Chinese/Indian takeaways (and restaurants when using their takeaway service) and foods from well-known fast food chain style restaurants such as McDonald's, Burger King, Dominos and Kentucky Fried Chicken (KFC). Please also include any takeaway meals which are delivered to your home from the establishment.

22. Do you eat takeaway or fast food? *Select one answer*

Yes (please carry on with questionnaire) ….. ❐1
No (please go to question 35 and carry on with questionnaire) …… ❐2

23. How often do you usually eat takeaway/fast food? *Select one answer*

Never or less than once/month……❐1
1-3 per month ……❐2
Once a week ……❐3
2-4 per week ……❐4
5-6 per week ……❐5
Once a day ……❐6
More than once a day ……❐7

24. What type of takeaway/fast food do you eat most often? *Please tick all that apply*

Chinese ……❐1
Indian ……❐2
Kebab ……❐3
Fried Chicken ……❐4
Pizza ……❐5
Fish and Chips ……❐6
Chips ……❐7
Beef Burger ……❐8
Other (Please specify)……❐9
………………………………………………………………………………………

Please complete the table below about the takeaway/fast food you eat
PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
25. Chinese Takeaway Meals ***(medium serving)***	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Beef with Green peppers in Black Bean Sauce	1	2	3	4	5	6	7	8	9
Crispy Chilli Beef	1	2	3	4	5	6	7	8	9
Special Chow Mein	1	2	3	4	5	6	7	8	9
Chicken Chow Mein	1	2	3	4	5	6	7	8	9
Char Sui Chow Mein	1	2	3	4	5	6	7	8	9
Kung Po King Prawns	1	2	3	4	5	6	7	8	9
Duck with Lemon, Orange or Plum Sauce	1	2	3	4	5	6	7	8	9

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
25. Chinese Takeaway Meals (continued) ***(medium serving)***	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Chicken Satay	1	2	3	4	5	6	7	8	9
Vegetable Curry	1	2	3	4	5	6	7	8	9
Sweet and Sour or Cantonese Style Chicken	1	2	3	4	5	6	7	8	9
Salt and Pepper Chicken	1	2	3	4	5	6	7	8	9
Salt and Pepper Chips	1	2	3	4	5	6	7	8	9
Chicken Wings	1	2	3	4	5	6	7	8	9
Spare Ribs	1	2	3	4	5	6	7	8	9
Crispy Duck Pancakes (1/4)	1	2	3	4	5	6	7	8	9
Noodles	1	2	3	4	5	6	7	8	9
Boiled Rice	1	2	3	4	5	6	7	8	9
Special Fried Rice/Yeung Chow Fried Rice	1	2	3	4	5	6	7	8	9
Egg Fried Rice	1	2	3	4	5	6	7	8	9
Spring Rolls	1	2	3	4	5	6	7	8	9
Prawn Crackers	1	2	3	4	5	6	7	8	9
Prawn Toast	1	2	3	4	5	6	7	8	9
Siu Mai (steamed meat dumplings)	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	**AVERAGE USE LAST YEAR**								
26. Indian Takeaway Meals *(medium serving)*	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Chicken Korma	1	2	3	4	5	6	7	8	9
Chicken Tikka Massala	1	2	3	4	5	6	7	8	9
Chicken Madras	1	2	3	4	5	6	7	8	9
King Prawn Rogan Josh	1	2	3	4	5	6	7	8	9
Lamb Rogan Josh	1	2	3	4	5	6	7	8	9
Lamb Bhuna	1	2	3	4	5	6	7	8	9
Vegetable Biryani	1	2	3	4	5	6	7	8	9
Pappadums with dipping sauces	1	2	3	4	5	6	7	8	9
Vegetable Samosa	1	2	3	4	5	6	7	8	9
Onion Bahji	1	2	3	4	5	6	7	8	9
Sag Paneer	1	2	3	4	5	6	7	8	9
Naan Bread	1	2	3	4	5	6	7	8	9
Pilau Rice	1	2	3	4	5	6	7	8	9
Boiled Rice (Basmati)	1	2	3	4	5	6	7	8	9
Keema Pilau Rice	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
27. English Takeaway Meals ***(medium serving)***	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Fish (Battered cod)	1	2	3	4	5	6	7	8	9
Scampi	1	2	3	4	5	6	7	8	9
Sausage	1	2	3	4	5	6	7	8	9
Chips	1	2	3	4	5	6	7	8	9
Chips and Curry Sauce	1	2	3	4	5	6	7	8	9
Chips and Gravy	1	2	3	4	5	6	7	8	9
Chips and Cheese	1	2	3	4	5	6	7	8	9
Omlette	1	2	3	4	5	6	7	8	9
28. Pizza Takeaway Meals including those from chain fast food e.g. Dominos and Pizza Hut ***(medium serving)***									
Margherita Pizza	1	2	3	4	5	6	7	8	9
Pepperoni Pizza	1	2	3	4	5	6	7	8	9
Seafood Pizza	1	2	3	4	5	6	7	8	9
Ham and Pineapple Pizza	1	2	3	4	5	6	7	8	9
Meat Pizza	1	2	3	4	5	6	7	8	9
Garlic Bread	1	2	3	4	5	6	7	8	9
Garlic Bread with Cheese	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please check that you have a tick (✓) on EVERY line

PLEASE PUT A TICK (✓) ON EVERY LINE.

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
29. Kebab Takeaway Meals *(medium serving)*	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Donner Kebab	1	2	3	4	5	6	7	8	9
Chicken Kebab	1	2	3	4	5	6	7	8	9
Sheesh Kebab	1	2	3	4	5	6	7	8	9
30. Fast Food Meals *(medium serving)*									
Fried Chicken	1	2	3	4	5	6	7	8	9
Beef Burger	1	2	3	4	5	6	7	8	9
Cheeseburger	1	2	3	4	5	6	7	8	9
Big Mac	1	2	3	4	5	6	7	8	9
Chicken Burger	1	2	3	4	5	6	7	8	9
Chicken Nuggets	1	2	3	4	5	6	7	8	9
Chicken Wrap with Mayo	1	2	3	4	5	6	7	8	9
Fries	1	2	3	4	5	6	7	8	9
McDonalds Breakfast	1	2	3	4	5	6	7	8	9
	1	2	3	4	5	6	7	8	9
Subway 12inch	1	2	3	4	5	6	7	8	9
	Never or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per 9

Please check that you have a tick (✓) on EVERY line

31. What particular takeaway food is your favourite?
Please give as much detail about the food or meal

………
………
………

32. Why do you eat takeaway foods?
Please tick all that apply

They are cheap ……❒1
They are easily available ……❒2
I don't like to prepare food ……❒3
I am usually too busy to cook ……❒4
I like the taste ……❒5
I don't know how to cook ……❒6
They are available close to my home ……❒7
For social occasions ……❒8
My friends and/or family like it ……❒9
Other (Please Specify)……❒10
………………………………………………………………

33. Where do you eat your takeaway food? *Please tick all that apply*

At home ……❒1
At a friend/families ……❒2
At the takeaway/fast food outlet ……❒3
On the street……❒4
Other (Please Specify)……❒5
………………………………………………………………

34. Who do you eat your takeaway or fast food with?
Please tick all that apply

Alone ……❒1
With family ……❒2
With friends ……❒3
Other (Please Specify)……❒4

About you and your health

For this section please try to be as accurate and honest as possible. The information you provide will be anonymised and stored securely so that it is inaccessible and unidentifiable.

35. What is your gender? *Select one answer*

Male ......❒1
Female ......❒2
Prefer not to say ......❒3

36. What is your age (Years)? *Select one answer*

18 - 25......❒1
26 - 35......❒2
36 - 45......❒3
46 - 55......❒4
56 - 65......❒5

37. What is your ethnicity? *Select one answer*

White British ......❒1
White Irish ......❒2
Other White ......❒3
Chinese ......❒4
Indian ......❒5
Pakistani ......❒6
Bangladeshi ......❒7
Other South Asian ......❒8
Black Caribbean ......❒9
Black African ......❒10
Black Other ......❒11
Mixed Race ......❒12
Other (Please Specify)......❒13

38. What is your marital status? *Select one answer*

Single ......❒1
Couple ......❒2
Married ......❒3
Civil Partnership ......❒4
Divorced ......❒5
Separated ......❒6
Widowed ......❒7
Other (Please Specify ......❒8
…………………………………………………..

39. What is your education? *Select one answer*

No formal qualifications ……❐1
GCSE/O-Level ……❐2
A-Level or Equivalent ……❐3
Degree Level ……❐4
Postgraduate Qualification ……❐5
Other (Please Specify)……❐6
………………………………………………

40. What is your employment status? *Select one answer*

Employed/self-employed ……❐1
Not in employment ……❐2
Full Time Student ……❐3
Retired ……❐4
Other (Please Specify)……❐5
……………………………………………………

41. What is your main occupation? *Select one answer*

Managers, Directors and Senior Officials ……❐1
Professional Occupations ……❐2
Associate Professional and Technical Occupations ……❐3
Administrative and Secretarial Occupations ……❐4
Skilled Trade Occupations ……❐5
Caring, Leisure and Other Service Occupations ……❐6
Sales and Customer Service Occupations ……❐7
Process, Plant and Machine Operatives ……❐8
Elementary Occupations ……❐9
Other (Please Specify) ……❐10
…………………………………………….

42. How many adults are in your household (18 years and over)? *Select one answer*

1……❐1
2……❐2
3……❐3
4 or more……❐4

43. Living arrangements * Who do you live with? *Select one answer*

On my own ……❐1
Wife/husband/partner ……❐2
Parents ……❐3
Friends ……❐4
Children ……❐5

44. How many children are in your household (under 18 years)? *Select one answer*

0……❒1
1……❒2
2……❒3
3……❒4
4 or more……❒5

45. Do you smoke? *Select one answer*

Yes, I smoke daily ……❒1
Yes, I smoke occasionally ……❒2
No, I used to smoke ……❒3
No, I have never smoked ……❒4

46. What is your current height? ____feet _____inches ***or*** _____cm

47. **What is your current weight?** ____stones _____pounds ***or*** **_____**kg

48. What is your current waist size? ____ inches ***or*** ____ cm

49. For your age, would you say that your health was:

Please tick one box on the scale of 1 to 5:

	1	2	3	4	5	
very good	❒	❒	❒	❒	❒	very poor

50. Which of the following best describes your daily work or other daytime activity that you usually do? *Select one answer*

I am usually sitting and do not walk about much (e.g. in an office)……❒1
I stand or walk about quite a lot, but do not have to carry or lift things very often (e.g. shop assistant, hairdresser)……❒2
I usually lift or carry light loads or have to climb stairs or hills often (e.g. plumber, electrician, carpenter)……❒3
I do heavy work or carry heavy loads often (e.g. dock worker, miner, bricklayer, construction worker)……❒4

51. **Please give the average number of hours per week you spend doing sports and other activities.**

Please write in the amount for each; if none write "0".

a) Mildly energetic
(e.g. walking, gardening, playing darts, general housework) _____ hour/w

b) Moderately energetic
(e.g. heavy housework or gardening, dancing, golf, cycling, leisurely swimming) _____ hour/w

c) Vigorous
(e.g. running, competitive swimming or cycling, tennis, football, squash, aerobics) _____ hour/w

52. **In a typical 7-day week, including the weekend, how many standard drinks of alcohol do you drink?** *(see the table below)*

Please write the number in the box below.

I usually drink ____ standard drinks of alcohol per week

ONE STANDARD DRINK = 1/2 pint of beer
or 1/2 pint cider
or 1/2 pint lager
or 1 glass of wine, martini, or cinzano
or 1 small glass of Sherry or Port
or 1 measure of Spirits (gin, whiskey, vodka etc.)
or 1 measure liquor

A PINT OF BEER, CIDER, OR LAGER COUNTS AS TWO STANDARD DRINKS

A DOUBLE MEASURE OF SPIRITS COUNTS AS TWO STANDARD DRINKS

53. **(a) Do you have any long-term illness, physical or mental health problem or handicap?**

Yes……❒1
No……❒2

54. **(b) If yes, does this limit your daily activity in any way?**

Yes……❒1
No……❒2

THANK YOU FOR TAKING TIME TO COMPLETE THIS QUESTIONNAIRE ☺

9.15 Study 1 Diet record - 24 hour ‘multiple-pass’ dietary recall

Diet History - 24 Hour Dietary recall

Participant Number: ………………….. Date: ………….. Day of the Week: …………………….

Is this a typical day? Yes ☐ No ☐

24 hour food record			
Time	**Details of Food and Drink (plus cooking method)**	**Portion/Quantity**	**Food Checklist**
			• Sweets • Chocolate • Biscuits • Cakes • Crisps • Nuts • Pies/Pastry • Cheese • Milk • Spread • Fried Foods • Fruit • Vegetables • Pulses • Oily Fish • Red Meat **Takeaway Foods** • Chinese • Indian • Kebab • Fried Chicken • Pizza • Fish and Chips • Chips • Beef Burger
Supplements:			
Alcohol:			
Additional Notes:			

9.16 Study 1 18 food item/groups (g/day) from 24 h MPR split by dietary pattern group from EHS

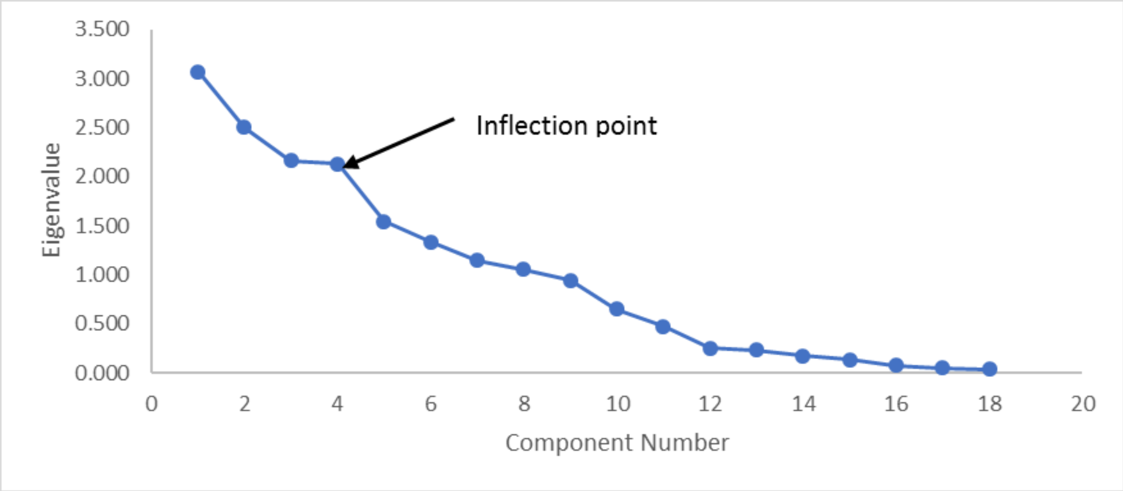
Food item/groups (g/day)	Traditional DP	Cosmopolitan DP	Convenience DP
Meat	119.83	58.86	98.40
Fish	27.22	92.50	22.67
Refined grains	110.47	117.43	146.59
Wholegrains	102.72	108.76	27.70
Potatoes	85.78	64.33	35.20
Dairy	252.78	210.33	203.40
Eggs	14.00	41.00	53.87
Fats and oils	23.69	31.45	21.67
Cakes, biscuits and crisps	48.68	25.48	38.51
Sweets and desserts	16.50	33.52	20.80
Sauces and condiments	30.14	9.38	17.87
Tea and Coffee	368.89	647.62	473.33
Alcohol	213.11	35.71	292.27
SSB	222.61	39.52	244.67
Fruit	121.44	110.67	90.87
Vegetables	119.78	323.33	156.87
Legumes	4.11	229.02	20.00
Takeaway food	69.34	0.00	80.67

NB: Highest amount (g/day) for each item are in red

9.17 Study 1 PCA correlation matrix

	Meat	Fish	Refined	Whole-	Potatoes	Dairy	Eggs	Fats and	Cakes	Sweet	Sauces	Tea	Alcohol	SSB	Fruit	Veg	Legumes	Takeaway
Meat	1.000	-.160	.191	-.239	.299	.399	.187	-.116	-.124	.302	-.395	.159	.272	-.199	-.125	-.180	-.446	.016
Fish	-.160	1.000	.026	.241	-.169	-.024	.193	-.121	-.335	-.123	-.131	.156	-.204	-.314	-.170	.275	.103	.187
Refined Grains	.191	.026	1.000	.141	.058	.264	.177	.309	.363	-.001	-.216	-.066	.077	.049	.186	.026	-.163	.460
Wholegrains	-.239	.241	.141	1.000	-.128	.200	-.043	-.370	.185	.352	-.115	-.229	.073	.245	.134	-.032	.300	-.228
Potatoes	.299	-.169	.058	-.128	1.000	.127	-.115	.262	.347	.399	-.182	-.067	.238	.444	-.237	-.199	-.194	.051
Dairy	.399	-.024	.264	.200	.127	1.000	.415	.112	.134	.489	-.114	.288	-.034	-.013	.189	-.149	-.464	.020
Eggs	.187	.193	.177	-.043	-.115	.415	1.000	-.162	-.266	-.003	.163	.045	-.175	-.051	.328	.542	-.033	.321
Fats and Oils	-.116	-.121	.309	-.370	.262	.112	-.162	1.000	.478	-.055	.203	.371	.142	-.004	-.075	-.019	-.237	.319
Cakes	-.124	-.335	.363	.185	.347	.134	-.266	.478	1.000	.075	.033	-.027	.120	.547	.045	-.141	-.156	.041
Sweet	.302	-.123	-.001	.352	.399	.489	-.003	-.055	.075	1.000	.044	.158	.195	-.028	.121	-.187	.014	-.199
Sauces	-.395	-.131	-.216	-.115	-.182	-.114	.163	.203	.033	.044	1.000	.105	-.082	-.117	.080	.094	.472	.222
Tea and Coffee	.159	.156	-.066	-.229	-.067	.288	.045	.371	-.027	.158	.105	1.000	-.111	-.503	-.204	.082	-.294	.066
Alcohol	.272	-.204	.077	.073	.238	-.034	-.175	.142	.120	.195	-.082	-.111	1.000	.002	-.216	-.171	-.058	-.284
SSB	-.199	-.314	.049	.245	.444	-.013	-.051	-.004	.547	-.028	-.117	-.503	.002	1.000	.079	-.038	.034	.035
Fruit	-.125	-.170	.186	.134	-.237	.189	.328	-.075	.045	.121	.080	-.204	-.216	.079	1.000	.372	.148	-.126
Veg	-.180	.275	.026	-.032	-.199	-.149	.542	-.019	-.141	-.187	.094	.082	-.171	-.038	.372	1.000	.392	.058
Legumes	-.446	.103	-.163	.300	-.194	-.464	-.033	-.237	-.156	.014	.472	-.294	-.058	.034	.148	.392	1.000	-.112
Takeaways	.016	.187	.460	-.228	.051	.020	.321	.319	.041	-.199	.222	.066	-.284	.035	-.126	.058	-.112	1.000

9.18 STUDY 1 PCA SCREE PLOT OF EIGENVALUES FOR EACH COMPONENT NUMBER



9.19 MPhil to PhD transfer (Approved 18th October 2016)

Reply Reply All Forward

Tue 18/10/2016 09:23

McKeon, Jo

Approval of Application to Transfer from MPhil to PhD

To Janssen, Hayley

Cc Stevenson, Leo; Davies, Ian; Richardson, Lucy

Dear Hayley,

I am very pleased to confirm that the Chair of the University's Research Degrees Committee has approved your application for transfer of registration from MPhil to PhD. This will be reported to RDC at its next scheduled meeting.

The Chair approved the application on behalf of RDC subject to the Independent Assessor's comments. These have been forwarded to your Director of Studies to enable you to receive feedback on your application.

Congratulations on your successful transfer from MPhil to PhD.

Kind regards,

LIVERPOOL JOHN MOORES UNIVERSITY

Jo McKeon
Research Student Support Officer,
Graduate School, Academic Registry,
4th Floor, Kingsway House Hatton Garden
Tel: 0151 904 6375 Fax: 0151 904 6462
Web: http://www2.ljmu.ac.uk/ Email: j.m.mck

9.20 STUDY 2 PARTICIPANT INFORMATION SHEET (ONLINE)

Participant information: Things you need to know before you start

Study Title: Takeaway eating habits in Merseyside

By H. Janssen (Faculty of Education, Health and Community, LJMU)

We invite you to complete a short survey about your eating habits. Before you decide to participate, please read the details below.

1. **What is the purpose of the study?** The aim of the survey is to investigate patterns of takeaway food consumption and factors that may influence consumption in the Merseyside area.
2. **Do I have to take part?** You are free to decide if you would like to take part or not and you can withdraw from the study at any time without giving a reason. To take part you should be **aged 18-64 years, eat takeaway/fast food** and **live in Liverpool, Knowsley, St.Helens, Sefton or the Wirral.**
3. **What will happen to me if I take part?** The 'Eating Habits Survey' survey takes approximately 20 minutes to complete and you can choose to stop at any time by saving to an e-mail address. The survey will ask questions about your eating habits and will ask you to provide details such as age group, gender, height and weight. You may also leave your contact details if you wish to be entered into a prize draw to **win a £200 Love2Shop voucher** or to take part in a further study. If you wish to leave your contact details you risk losing anonymity, however, only the researcher will know that you have taken part in the research and you will only be contacted to be invited to take part in further research or to inform you that you have won the prize draw.
4. **Will my taking part in the study be kept confidential?** Yes, all data collected about you during the course of the research will be kept strictly confidential.
5. **Are there any risks/benefits involved?** No risks. Fully completed surveys will be eligible to be entered into a prize draw to **win a £200 Love2Shop voucher**.
6. **Contact Details of Researcher**

If you have any questions about the study or about taking part please contact: Hayley Janssen, e-mail: **H.G.Janssen@2012.ljmu.ac.uk**

This study has received ethical approval from LJMU's Research Ethics Committee (16/EHC/002). *If you any concerns regarding your involvement in this research, please discuss these with the researcher in the first instance. If you wish to make a complaint, please contact researchethics@ljmu.ac.uk and your communication will be re-directed to an independent person as appropriate.*

For future reference you can download and save the participant information sheet here;
https://www.dropbox.com/s/khjkub3msj0djuf/Online%20Participant%20Information%20Sheet%20V5%2012.10.16.docx?dl=0

I understand that by completing this survey I am consenting to be part of this research study Yes, ☐ take me to the survey

9.21 STUDY 2 SOCIAL MEDIA CAMPAIGNS

Social Media Recruitment Campaigns

Social media (Facebook, Twitter etc.) campaigns will be used as a recruitment technique for the study. Other social media (Instagram, linked in) campaigns may be used depending on uptake. Participants will have access to a hyperlink to the participant information sheet on the social media message to read and download prior to taking part in the study. At the end of the participant information sheet there will be a hyperlink to the online survey. At the end of the online survey there will be a section inviting participants to provide contact details for the possibility of taking part in other parts of the study (dietary recalls and urine sample collection). For those who volunteer to take part, a cross check, on the telephone, will occur to ensure they have read and understood the participation sheet. Additional verbal consent, via a recorded telephone call, will be obtained from those who cannot provide written consent. The social media campaigns will include a brief description, an image of the recruitment flyer and a hyperlink to the participant information sheet as follows;

Twitter Campaign

Are you aged 18-64 and live in Liverpool? If so, click here to find out more and take part in this study: **https://ljmu.ac.uk/dietary-patterns-study-information**

Please re-post and share ☺

Facebook Campaign

Are you aged 18-64 and live in Liverpool? If so, why not take part in this study by LJMU about the food you eat. There is £600 in high street vouchers up for grabs… Click here to find out more and take part:

https://www.dropbox.com/s/fk4hmi4s57m90kh/Online%20Participant%20Information%20Sheet%20V3%2023.05.16.docx?dl=0

OR **https://drive.google.com/file/d/0B0F2-sL-gtbFdXhjQzVHZ2Q4WDA/view?usp=sharing** Please re-post and share ☺

9.22 Study 2 Flyer (final version takeaways)

9.23 Study 2 Poster with tear off invites

LIVERPOOL JOHN MOORES UNIVERSITY

PRIFYSGOL ABERYSTWYTH UNIVERSITY

PARTICIPANTS NEEDED!

THIS STUDY IS RESEARCHING THE PATTERNS OF TAKEAWAY FOOD CONSUMPTION IN MERSEYSIDE

EVER EAT TAKEAWAY OR FAST FOOD?

WE NEED VOLUNTEERS WHO:

LIVE IN MERSEYSIDE

ARE AGED 18-64 YEARS

FOLLOW THE LINK BELOW (OR SCAN QR CODE), FILL IN THE SURVEY AND BE ENTERED INTO A PRIZE DRAW...

https://ljmu.onlinesurveys.ac.uk/eating-habits-survey

WOULD YOU LIKE A CHANCE TO WIN £200 LOVE2SHOP VOUCHERS?

We would like to invite some people to a further study. If you are interested and would like more information please contact: H.G.Janssen@2012.ljmu.ac.uk You may withdraw from the study at any time. All information will stay confidential.

https://ljmu.onlinesurveys.ac.uk/ eating-habits-survey

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9.24 Study 2 Eating Habits Survey (on Online Surveys)

LIVERPOOL JOHN MOORES UNIVERSITY

Eating Habits Survey

About the food you eat

1. YOUR DIET IN THE PAST YEAR

How often you have eaten the following food items **during the past year.** Please estimate as best you can and tick ✓ a box for each food item ☺

If you have not eaten the food item please tick ✓ the NEVER box

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
MEAT & FISH ***(medium serving)*** ***(not fast food or takeaway)***	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Beef: roast, steak, mince, stew casserole, curry or bolognese	1	2	3	4	5	6	7	8	9
Beef burgers (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
Pork: roast, chops, stew, slice or curry	1	2	3	4	5	6	7	8	9
Lamb: roast, chops, stew or curry	1	2	3	4	5	6	7	8	9
Chicken, turkey or other poultry: including fried, casseroles or curry	1	2	3	4	5	6	7	8	9
Bacon	1	2	3	4	5	6	7	8	9
Ham	1	2	3	4	5	6	7	8	9
Corned beef, Spam, luncheon meats	1	2	3	4	5	6	7	8	9
Sausages	1	2	3	4	5	6	7	8	9
Savoury pies, e.g. meat pie, pork pie, pasties, steak & kidney pie, sausage rolls, scotch egg	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
MEAT & FISH,(continued) ***(medium serving)*** ***(not fast food or takeaway)***	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Liver, liver pate, liver sausage	1	2	3	4	5	6	7	8	9
Fried fish in batter, as in fish and chips (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
Fish fingers, fish cakes	1	2	3	4	5	6	7	8	9
Other white fish, fresh or frozen, e.g. cod, haddock, plaice, sole, halibut	1	2	3	4	5	6	7	8	9
Oily fish, fresh or canned, e.g. mackerel, kippers, tuna, salmon, sardines, herring	1	2	3	4	5	6	7	8	9
Shellfish, e.g. crab, prawns, mussels	1	2	3	4	5	6	7	8	9
BREAD & SAVOURY BISCUITS ***(one slice or biscuit)*** ***(not fast food or takeaway)***									
White bread and rolls	1	2	3	4	5	6	7	8	9
Scones, teacakes, crumpets, muffins or croissants	1	2	3	4	5	6	7	8	9
Brown bread and rolls	1	2	3	4	5	6	7	8	9
Wholemeal bread and rolls	1	2	3	4	5	6	7	8	9
Cream crackers, cheese biscuits	1	2	3	4	5	6	7	8	9
Pitta bread, naan bread, chapatti (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
Garlic bread (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
CEREALS ***(one bowl)***	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Porridge, Readybrek	1	2	3	4	5	6	7	8	9
Sugar coated cereals e.g. Sugar Puffs, Cocoa Pops, Frosties	1	2	3	4	5	6	7	8	9
Non-sugar coated cereals e.g. Cornflakes, Rice Crispies	1	2	3	4	5	6	7	8	9
All Bran, Bran Flakes, Muesli	1	2	3	4	5	6	7	8	9
Wholegrain cereals e.g. Cheerios, Weetabix, Shredded Wheat	1	2	3	4	5	6	7	8	9
POTATOES, RICE & PASTA ***(medium serving)*** ***(not fast food or takeaway)***									
Boiled, mashed, instant or jacket potatoes	1	2	3	4	5	6	7	8	9
Chips, potato waffles, potato wedges (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
Roast potatoes	1	2	3	4	5	6	7	8	9
Yorkshire pudding, pancakes, dumpling	1	2	3	4	5	6	7	8	9
Potato salad	1	2	3	4	5	6	7	8	9
White rice	1	2	3	4	5	6	7	8	9
Brown rice	1	2	3	4	5	6	7	8	9
White or green pasta, e.g. spaghetti, macaroni, noodles	1	2	3	4	5	6	7	8	9
Tinned pasta, e.g. spaghetti, ravioli, macaroni	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
POTATOES, RICE & PASTA (continued) ***(medium serving)*** ***(not fast food or takeaway)***	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Super noodles, pot noodles, pot savouries	1	2	3	4	5	6	7	8	9
Wholemeal pasta	1	2	3	4	5	6	7	8	9
Lasagne, moussaka, cannelloni	1	2	3	4	5	6	7	8	9
Pizza (not fast food or takeaway)	1	2	3	4	5	6	7	8	9
DAIRY PRODUCTS & FATS									
Single or sour cream *(tablespoon)*	1	2	3	4	5	6	7	8	9
Double or clotted cream *(tablespoon)*	1	2	3	4	5	6	7	8	9
Low fat yoghurt, low fat greek yoghurt, fromage frais *(125g carton)*	1	2	3	4	5	6	7	8	9
Full fat or Greek yoghurt *(125g carton)*	1	2	3	4	5	6	7	8	9
Dairy desserts *(125g carton),* e.g. mousse or custard	1	2	3	4	5	6	7	8	9
Cheese, e.g. Cheddar, Brie, Edam *(medium serving)*	1	2	3	4	5	6	7	8	9
Cottage cheese, low fat soft cheese *(medium serving)*	1	2	3	4	5	6	7	8	9
Eggs as boiled, fried, scrambled, poached, omelette etc. *(one)*	1	2	3	4	5	6	7	8	9
Quiche *(medium serving)*	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
The following DAIRY/FATS on bread or vegetables *(teaspoon)*	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Butter	1	2	3	4	5	6	7	8	9
Block margarine, e.g. Stork, Krona	1	2	3	4	5	6	7	8	9
Polyunsaturated margarine, e.g. Flora sunflower	1	2	3	4	5	6	7	8	9
Other soft margarine, dairy spreads, e.g. Blue Band, Clover	1	2	3	4	5	6	7	8	9
Low fat spread, e.g. Gold	1	2	3	4	5	6	7	8	9
SWEETS & SNACKS									
Sweet biscuits, chocolate, e.g. digestive *(one)*	1	2	3	4	5	6	7	8	9
Sweet biscuits, plain, e.g. Nice, ginger *(one)*	1	2	3	4	5	6	7	8	9
Cakes e.g. fruit, sponge, sponge pudding *(medium serving)*	1	2	3	4	5	6	7	8	9
Sweet buns & pastries e.g. flapjacks, doughnuts, Danish pastries, cream cakes *(medium serving)*	1	2	3	4	5	6	7	8	9
Fruit pies, tarts, crumbles *(medium serving)*	1	2	3	4	5	6	7	8	9
Milk puddings, e.g. rice, custard, trifle *(medium serving)*	1	2	3	4	5	6	7	8	9
Ice cream, choc ices *(one)*	1	2	3	4	5	6	7	8	9
Chocolates *(small bar or ¼ pound of chocolates)*	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
SWEETS & SNACKS (continued)	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Sweets, toffees, mints *(one packet)*	1	2	3	4	5	6	7	8	9
Sugar added to tea, coffee, cereal *(teaspoon)*	1	2	3	4	5	6	7	8	9
Crisps or other packet snacks e.g. Wotsits *(one packet)*	1	2	3	4	5	6	7	8	9
Peanuts or other nuts *(one packet)*	1	2	3	4	5	6	7	8	9
SOUPS, SAUCES AND SPREADS									
Vegetable soups *(bowl)*	1	2	3	4	5	6	7	8	9
Meat soups *(bowl)*	1	2	3	4	5	6	7	8	9
Sauces, e.g. white sauce, cheese sauce, gravy *(medium serving)*	1	2	3	4	5	6	7	8	9
Tomato based sauces e.g. pasta sauces *(medium serving)*	1	2	3	4	5	6	7	8	9
Tomato ketchup, brown sauce *(tablespoon)*	1	2	3	4	5	6	7	8	9
Relishes e. g. pickles, chutney, mustard *(tablespoon)*	1	2	3	4	5	6	7	8	9
Low calorie, low fat salad cream or mayonnaise *(tablespoon)*	1	2	3	4	5	6	7	8	9
Salad cream, mayonnaise *(tablespoon)*	1	2	3	4	5	6	7	8	9
French dressing *(tablespoon)*	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
SOUPS, SAUCES AND SPREADS (continued)	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Other salad dressing *(tablespoon)*	1	2	3	4	5	6	7	8	9
Marmite, Bovril *(teaspoon)*	1	2	3	4	5	6	7	8	9
Jam, marmalade, honey, syrup *(teaspoon)*	1	2	3	4	5	6	7	8	9
Peanut butter *(teaspoon)*	1	2	3	4	5	6	7	8	9
Chocolate spread, chocolate nut spread e.g. nutella *(teaspoon)*	1	2	3	4	5	6	7	8	9
Dips e.g. hummus, cheese and chive *(tablespoon)*	1	2	3	4	5	6	7	8	9
DRINKS									
Tea *(cup)*	1	2	3	4	5	6	7	8	9
Coffee, instant or ground *(cup)*	1	2	3	4	5	6	7	8	9
Coffee whitener, e.g. Coffee-mate *(teaspoon)*	1	2	3	4	5	6	7	8	9
Cocoa, hot chocolate *(cup)*	1	2	3	4	5	6	7	8	9
Horlicks, Ovaltine *(cup)*	1	2	3	4	5	6	7	8	9
Wine *(glass)*	1	2	3	4	5	6	7	8	9
Beer, lager or cider *(half pint)*	1	2	3	4	5	6	7	8	9
Port, sherry, vermouth, liqueurs *(glass)*	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
DRINKS (continued)	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Spirits, e.g. gin, brandy, whisky, vodka *(single)*	1	2	3	4	5	6	7	8	9
Low calorie or diet fizzy soft drinks *(glass)*	1	2	3	4	5	6	7	8	9
Fizzy soft drinks, e.g. Coca cola, lemonade *(glass)*	1	2	3	4	5	6	7	8	9
Pure fruit juice (100%) e.g. orange, apple juice *(glass)*	1	2	3	4	5	6	7	8	9
Fruit squash or cordial *(glass)*	1	2	3	4	5	6	7	8	9
FRUIT *(1 fruit or medium serving)* *For very seasonal fruits such as strawberries, please estimate your average use when the fruit is in season									
Apples	1	2	3	4	5	6	7	8	9
Pears	1	2	3	4	5	6	7	8	9
Oranges, satsumas, mandarins, tangerines, clementines	1	2	3	4	5	6	7	8	9
Grapefruit	1	2	3	4	5	6	7	8	9
Bananas	1	2	3	4	5	6	7	8	9
Grapes	1	2	3	4	5	6	7	8	9
Melon	1	2	3	4	5	6	7	8	9
*Peaches, plums, apricots, nectarines	1	2	3	4	5	6	7	8	9
*Strawberries, raspberries, kiwi fruit	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
FRUIT (continued) ***(1 fruit or medium serving)***	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Tinned fruit	1	2	3	4	5	6	7	8	9
Dried fruit, e.g. raisins, prunes, figs	1	2	3	4	5	6	7	8	9
VEGETABLES Fresh, frozen or tinned ***(medium serving)***									
Carrots	1	2	3	4	5	6	7	8	9
Spinach	1	2	3	4	5	6	7	8	9
Broccoli	1	2	3	4	5	6	7	8	9
Brussels sprouts	1	2	3	4	5	6	7	8	9
Cabbage	1	2	3	4	5	6	7	8	9
Peas	1	2	3	4	5	6	7	8	9
Green beans, broad beans, runner beans	1	2	3	4	5	6	7	8	9
Marrow, courgettes	1	2	3	4	5	6	7	8	9
Cauliflower	1	2	3	4	5	6	7	8	9
Parsnips, turnips, swedes	1	2	3	4	5	6	7	8	9
Leeks	1	2	3	4	5	6	7	8	9
Onions	1	2	3	4	5	6	7	8	9
Garlic	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ☑ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
VEGETABLES **Fresh, frozen or tinned (continued)** ***(medium serving)***	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Mushrooms	1	2	3	4	5	6	7	8	9
Sweet peppers	1	2	3	4	5	6	7	8	9
Beansprouts	1	2	3	4	5	6	7	8	9
Green salad, lettuce, cucumber, celery	1	2	3	4	5	6	7	8	9
Mixed vegetables (frozen or tinned)	1	2	3	4	5	6	7	8	9
Watercress	1	2	3	4	5	6	7	8	9
Tomatoes	1	2	3	4	5	6	7	8	9
Sweetcorn	1	2	3	4	5	6	7	8	9
Beetroot, radishes	1	2	3	4	5	6	7	8	9
Coleslaw	1	2	3	4	5	6	7	8	9
Avocado	1	2	3	4	5	6	7	8	9
Baked Beans	1	2	3	4	5	6	7	8	9
Dried lentils, beans, peas	1	2	3	4	5	6	7	8	9
Tofu, soya meat, TVP, Vegeburger	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

YOUR DIET IN THE PAST YEAR, CONTINUED

2. **Were there any OTHER foods that you ate more than once a week?**

Yes...... ☐1
No...... ☐2

If **YES**, please list below

Food	Usual serving size	Number of times eaten each week

3. **What type of milk did you most often use?** *Select one answer*

Full cream (blue top)...... ☐1
Channel Islands...... ☐2
Dried milk...... ☐3
Semi-skimmed (green top)...... ☐4
Skimmed (red top) ...... ☐5
Soya...... ☐6
None ...... ☐7
Other (Please Specify)...... ☐8

....

4. **Approximately, how much milk did you drink EACH DAY, including milk with tea, coffee, cereals etc?** *Select one answer*

None ...... ☐1
Quarter of a pint (roughly 125mls) ...... ☐2
Half a pint (roughly 250mls) ...... ☐3
Three quarters of a pint (roughly 375mls) ...... ☐4
One pint (roughly 500mls) ...... ☐5
More than one pint (more than 500mls) ...... ☐6

5. **What kind of fat did you most often use for frying, roasting, grilling etc?**
Select one answer

Butter……☐1
Lard/dripping……☐2
Solid vegetable fat……☐3
Margarine……☐4
Vegetable oil……☐5
Olive oil……☐6
None……☐7
Other (Please Specify)…… ☐8
……………………………………………………………………………………

6. **Have you taken any Vitamins, Minerals, Fish Oils or other Food Supplements during the past year?**

Yes……☐1
No……☐2
Not Sure……☐3

7. **During the course of last year, on average, how many times a week did you eat the following foods?**

Food Type	Times/week	Portion size
Vegetables (not including potatoes)	☐	medium serving
Salads	☐	medium serving
Fruit and fruit products (not Including fruit	☐	medium serving juice) or 1 fruit
Fish and fish product	☐	medium serving
Meat, meat products and meat dishes (including bacon, ham and chicken)	☐	medium serving

8. TAKEAWAY/FAST FOOD EATEN IN THE PAST YEAR

Food from **takeaway outlets** such as **Chinese, Kebab, Chippy and Indian** (and restaurants when using their takeaway service) and foods from **fast food restaurants** such as **McDonald's, Burger King, Dominos and Kentucky Fried Chicken (KFC).** Please also include takeaway meals delivered to your home.

How often you have eaten the following food items **during the past year.** Please estimate as best you can and tick ✓ a box for each food item ☺

If you have not eaten the food item please tick ✓ the NEVER box

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
FAST FOOD MEALS ***(medium serving)***	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Beef Burger, e.g. Big Mac, Whopper, Big King, Cheeseburger, Hamburger	1	2	3	4	5	6	7	8	9
Chicken Burger, Chicken Sandwich, Chicken Royale, Chicken Wrap	1	2	3	4	5	6	7	8	9
Fried Chicken, e.g. Wings, Breast, Nuggets	1	2	3	4	5	6	7	8	9
Fish Burger, e.g. Fish Filet Burger, King Fish	1	2	3	4	5	6	7	8	9
Veggie Burger, e.g. Veggie Bean Burger, Veggie Wrap	1	2	3	4	5	6	7	8	9
Mozzarella Sticks, Onion Rings, Cheesy Bites	1	2	3	4	5	6	7	8	9
Fries, Wedges, Curly Fries, Hash Browns	1	2	3	4	5	6	7	8	9
McDonalds Breakfast, e.g. Muffin, Pancake Breakfast, Bacon roll	1	2	3	4	5	6	7	8	9
Subway 12inch, e.g. Meatball Marinara, B.L.T, Chicken Teriyaki	1	2	3	4	5	6	7	8	9
Sauces or Dips, e.g. Tomato Ketchup, BBQ, Sweet Chilli, Mayo	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
CHINESE TAKEAWAY MEALS ***(medium serving)***	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Chicken Wings, e.g. Peking Style, Salt and Pepper, Honey Roast	1	2	3	4	5	6	7	8	9
Spare Ribs, e.g. BBQ, Salt and Pepper, Honey Roast	1	2	3	4	5	6	7	8	9
Crispy Duck Pancakes (1/4)	1	2	3	4	5	6	7	8	9
Spring Rolls, Crispy Won Tons, Prawn Toast	1	2	3	4	5	6	7	8	9
Prawn Crackers	1	2	3	4	5	6	7	8	9
Siu Mai, Dim Sum (steamed meat dumplings)	1	2	3	4	5	6	7	8	9
Skewered Chicken (with Satay or BBQ sauce)	1	2	3	4	5	6	7	8	9
Soup, e.g. Hot and Sour Soup, Chicken and Sweetcorn Soup, Won Ton Soup	1	2	3	4	5	6	7	8	9
Deep Fried Chicken with Salt and Pepper (Spicy)	1	2	3	4	5	6	7	8	9
Peking Style Dish, e.g. Shredded Chicken, Shredded Beef	1	2	3	4	5	6	7	8	9
Lemon, Orange or Plum Sauce Dish, e.g. Duck or Chicken	1	2	3	4	5	6	7	8	9
Satay Dish, e.g. Beef, Chicken	1	2	3	4	5	6	7	8	9
Sweet and Sour Dish, e.g. Chicken, Pork, King Prawn	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
CHINESE TAKEAWAY MEALS (continued) ***(medium serving)***	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Curry Dish e.g. Beef, Chicken, King Prawn, Special, Vegetable	1	2	3	4	5	6	7	8	9
Kung Po Dish, e.g. Beef, Chicken, King Prawn, Special, Vegetable	1	2	3	4	5	6	7	8	9
Foo Yung Dish, e.g. Special, Beef, Mushroom, King Prawn	1	2	3	4	5	6	7	8	9
Chop Suey Dish, e.g. Beef, Chicken, King Prawn, Special, Vegetable	1	2	3	4	5	6	7	8	9
Chow Mein Dish, e.g. Chicken, Special, Beef, Char sui, Vegetable	1	2	3	4	5	6	7	8	9
Beef Dish, e.g. Crispy Chilli Beef, Beef with green peppers in Black Bean Sauce, Chilli and Garlic Sauce	1	2	3	4	5	6	7	8	9
Chicken or Duck Dish, e.g. with Cashew Nuts, in Oyster sauce, in Black Bean Sauce, with mushrooms	1	2	3	4	5	6	7	8	9
Vegetarian Dish, e.g. with Cashew Nuts, Chinese Greens	1	2	3	4	5	6	7	8	9
King Prawn Dish, e.g. Chilli and Garlic Sauce, in Oyster sauce, in Black Bean Sauce, with mushrooms	1	2	3	4	5	6	7	8	9
Fried Rice Dish, e.g. Special, Yeung Chow, Chicken, Egg, Mushroom	1	2	3	4	5	6	7	8	9
Salt and Pepper Chips (If selecting chips find under English Takeaway Section)	1	2	3	4	5	6	7	8	9
Noodles and Beansprouts	1	2	3	4	5	6	7	8	9
Boiled Rice	1	2	3	4	5	6	7	8	9
Sauces, e.g. Sweet and Sour, Ok, Satay, Curry	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
INDIAN TAKEAWAY MEALS ***(medium serving)***	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Pappadums with dipping sauces	1	2	3	4	5	6	7	8	9
Samosa, Bhaji, Pakora	1	2	3	4	5	6	7	8	9
Chicken or Lamb Tikka Starter	1	2	3	4	5	6	7	8	9
Tandoori Chicken/King Prawn/Lamb	1	2	3	4	5	6	7	8	9
Korma Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Tikka Massala Dish or Massala Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Rogan Josh Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Bhuna Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Biryani Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Balti Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Sagwala Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Tikka Jalfrazi Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Dupiaza Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Medium Curry Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
INDIAN TAKEAWAY MEALS ***(medium serving)*** ***(continued)***	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Achari Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Vindaloo Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Pathia Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Pasanda Dish, e.g. Chicken, Lamb, King Prawn, Vegetable	1	2	3	4	5	6	7	8	9
Special Curry	1	2	3	4	5	6	7	8	9
Butter Chicken	1	2	3	4	5	6	7	8	9
Saag, e.g. Paneer, Aloo, Channa	1	2	3	4	5	6	7	8	9
Dal, e.g. Tarka	1	2	3	4	5	6	7	8	9
Naan Bread, e.g Garlic and Coriander Naan, Peshwari Naan, Cheese Naan, Keema Naan	1	2	3	4	5	6	7	8	9
Paratha, Chapati, e.g. Aloo, Keema	1	2	3	4	5	6	7	8	9
Pilau Rice	1	2	3	4	5	6	7	8	9
Fried Rice, e.g. Plain, Chicken, Keema	1	2	3	4	5	6	7	8	9
Boiled Rice (Basmati)	1	2	3	4	5	6	7	8	9
Sauces, e.g. Tikka Massala Sauce,	1	2	3	4	5	6	7	8	9
	NEVER or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day

Please estimate as best you can and tick ✓ a box for each food item ☺

FOODS & AMOUNTS	AVERAGE USE LAST YEAR								
ENGLISH TAKEAWAY MEALS *(medium serving)*	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Fish, e.g. Cod	1	2	3	4	5	6	7	8	9
Scampi	1	2	3	4	5	6	7	8	9
Sausage	1	2	3	4	5	6	7	8	9
Omlette	1	2	3	4	5	6	7	8	9
Roast Chicken	1	2	3	4	5	6	7	8	9
Chips	1	2	3	4	5	6	7	8	9
Sauces or Sides, e.g. Curry Sauce, Gravy, Mushy Peas, Cheese	1	2	3	4	5	6	7	8	9
PIZZA TAKEAWAY MEALS including those from chain fast food e.g. Dominos and Pizza Hut *(medium serving)*									
Cheese Pizza, e.g. Margherita, Four Cheese	1	2	3	4	5	6	7	8	9
Meat Pizza, e.g. meat feast, peperoni, spicy chicken, Ham and Pineapple, BBQ chicken, Philly Steak	1	2	3	4	5	6	7	8	9
Seafood/Fish Pizza, e.g. mixed seafood, tuna and sweetcorn, smoked salmon	1	2	3	4	5	6	7	8	9
Vegetarian Pizza, e.g. mixed vegetables, veggie hot one, spinach and ricotta	1	2	3	4	5	6	7	8	9
Garlic Bread or Garlic Bread with Cheese	1	2	3	4	5	6	7	8	9
KEBAB TAKEAWAY MEALS *(medium serving)*	**NEVER** or less than once/ month	1-3 per month	Once a week	2-4 per week	5-6 per week	Once a day	2-3 per day	4-5 per day	6+ per day
Donner Kebab	1	2	3	4	5	6	7	8	9
Chicken Kebab	1	2	3	4	5	6	7	8	9
Sheesh Kebab	1	2	3	4	5	6	7	8	9

9. Were there any OTHER takeaway/fast foods that you ate more than once a week?

Yes......☐1
No......☐2

If **YES**, please list below

Takeaway/fast food	Usual serving size	Number of times eaten each week

10. How often do you usually eat takeaway/fast food? *Select one answer*

Never or less than once/month......☐1
1-3 per month ......☐2
Once a week ......☐3
2-4 per week ......☐4
5-6 per week ......☐5
Once a day ......☐6
More than once a day ......☐7

11. Why do you eat takeaway/fast foods?

Please tick all that apply

I have not eaten takeaway/fast food in the past year ......☐1
They are cheap ......☐2
They are easily available ......☐3
I don't like to prepare food ......☐4
I am usually too busy to cook ......☐5
I like the taste ......☐6
I don't know how to cook ......☐7
They are available close to my home ......☐8
For social occasions ......☐9

My friends and/or family like it ......☐10
Other (Please Specify)...... ☐11
....

12. **Where do you eat your takeaway/fast food?**
Please tick all that apply
I have not eaten takeaway/fast food in the past year ......☐1
At home ......☐2
At a friend/families ......☐3
At the takeaway/fast food outlet ......☐4
On the street......☐5
Other (Please Specify)...... ☐6
....

13. Who do you eat your takeaway/fast food with?
Please tick all that apply
I have not eaten takeaway/fast food in the past year ......☐1
Alone ......☐2
With family ......☐3
With friends ......☐4
Other (Please Specify)...... ☐5
....

About you and your background

For this section please try to be as accurate and honest as possible. The information you provide will be anonymised and stored securely so that it is inaccessible and unidentifiable.

14. What is your gender? *Select one answer*

Male ......☐1
Female ......☐2
Transgender ......☐3

15. What is your age group (Years)? *Select one answer*

18 - 25......☐1
26 - 35......☐2
36 - 45......☐3
46 - 55......☐4
56 - 64......☐5

16. What is your ethnicity? *Select one answer*

White British ......☐1
White Irish ......☐2
Other White ......☐3
Chinese ......☐4
Indian ......☐5
Pakistani ......☐6
Bangladeshi ......☐7
Other South Asian ......☐8
Black Caribbean ......☐9
Black African ......☐10
Black Other ......☐11
Mixed Race ......☐12
Other (Please Specify)...... ☐13

....

17. What is your marital status? *Select one answer*

Single ......☐1
Married ......☐2
Civil Partnership ......☐3
Divorced ......☐4
Separated ......☐5
Widowed ......☐6
Other (Please Specify ......☐7

....

18. Living arrangements * Who do you live with? *Select one answer*

On my own ......☐1
Wife/husband/partner ......☐2
Parents ......☐3
Friends ......☐4
Children ......☐5
Other (Please Specify) ......☐6

....

19. How many adults are in your household (18 years and over)?
Select one answer

1......☐1
2......☐2
3......☐3
4 or more......☐4

20. How many children are in your household (under 18 years)?
Select one answer

0......☐1
1......☐2
2......☐3
3......☐4
4 or more......☐5

21. What is your postcode? *First section only*

……………………………………………………………………………

22. What is your highest qualification gained? *Select one answer*

No formal qualifications …… ☐ 1

GCSE/O-Level …… ☐ 2

A-Level or Equivalent …… ☐ 3

Degree Level …… ☐ 4

Postgraduate Qualification …… ☐ 5

Other (Please Specify) …… ☐ 6

……………………………………………………………………………

23. What is your employment status? *Select one answer*

Employed/self-employed …… ☐ 1

Not in employment …… ☐ 2

Full Time Student …… ☐ 3

Retired …… ☐ 4

Other (Please Specify)…… ☐ 5

……………………………………………………………………………

24. What is your main occupation? *Select one answer*

Managers, Directors and Senior Officials …… ☐ 1

Professional Occupations …… ☐ 2

Associate Professional and Technical Occupations …… ☐ 3

Administrative and Secretarial Occupations …… ☐ 4

Skilled Trade Occupations …… ☐ 5

Caring, Leisure and Other Service Occupations …… ☐ 6

Sales and Customer Service Occupations …… ☐ 7

Process, Plant and Machine Operatives …… ☐ 8

Elementary Occupations …… ☐ 9

N/A (student/unemployed/retired)…… ☐ 10

Other (Please Specify) …… ☐ 11

………………………………………………………………………………….

25. For your age, would you say that your health was:

Please tick one box on the scale of 1 to 5:

	1	2	3	4	5	
very good	☐	☐	☐	☐	☐	very poor

26. **Which of the following best describes your daily work or other daytime activity that you usually do?** *Select one answer*

I am usually sitting and do not walk about much (e.g. in an office)…… ☐ 1

I stand or walk about quite a lot, but do not have to carry or lift things very often (e.g. shop assistant, hairdresser)…… ☐ 2

I usually lift or carry light loads or have to climb stairs or hills often (e.g. plumber, electrician, carpenter)…… ☐ 3

I do heavy work or carry heavy loads often (e.g. dock worker, miner, bricklayer, construction worker)…… ☐ 4

27. Please give the average number of hours per week you spend doing SPORTS and OTHER activities (not your daily work):

Please write in the amount for each; if none write "0".

a) Mildly energetic (e.g. walking, gardening, playing darts, general housework)	_____ hour/w
b) Moderately energetic (e.g. heavy housework or gardening, dancing, golf, cycling, leisurely swimming)	_____ hour/w
c) Vigorous (e.g. running, competitive swimming or cycling, tennis, football, squash, aerobics)	_____ hour/w

28. What is your current height? ____feet _____inches ***or*** _____cm

29. What is your current weight? ____stones _____pounds ***or*** _____kg

30. What is your current waist size? ____ inches ***or*** ____ UK size

31. Do you smoke? *Select one answer*

Yes, I smoke daily ……☐1
Yes, I smoke occasionally ……☐2
No, I used to smoke ……☐3
No, I have never smoked ……☐4

32. **In a typical 7-day week, including the weekend, how many standard drinks of alcohol do you drink?** *(see the table below for more information)*

Please write the number in the box below.

I usually drink ______ standard drinks of alcohol per week

I do not drink alcohol ……☐1

ONE STANDARD DRINK =1/2 pint of beer
or 1/2 pint cider
or 1/2 pint lager
or 1 small glass of wine, martini, or cinzano
or 1 small glass of Sherry or Port
or 1 measure of Spirits (gin, whiskey, vodka...)
or 1 measure liquor

A PINT OF BEER, CIDER OR LAGER COUNTS AS TWO STANDARD DRINKS
A DOUBLE MEASURE OF SPIRITS COUNTS AS TWO STANDARD DRINKS

33.(a) Do you have any long-term illness, physical or mental health problem or handicap?

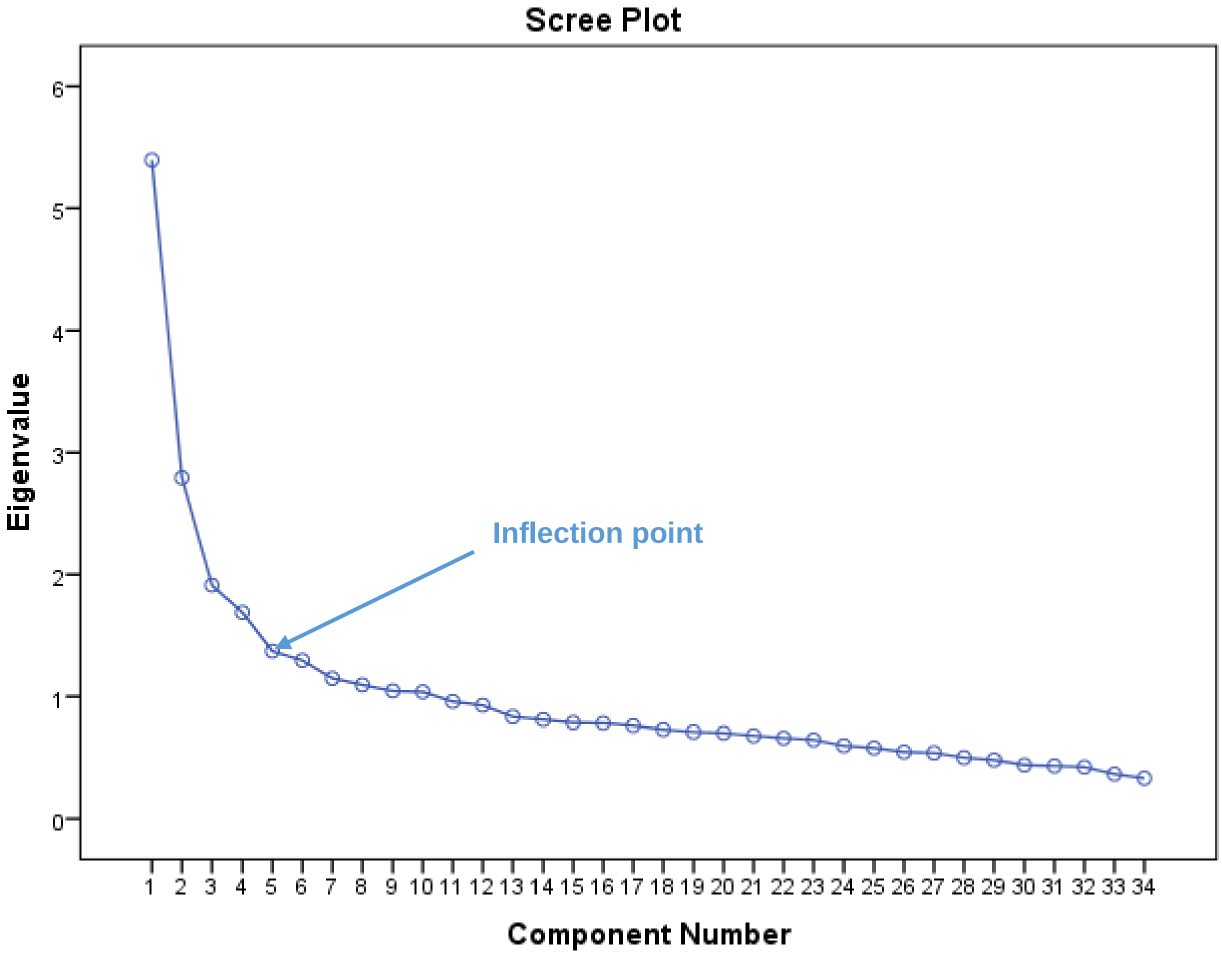
Yes……☐1
No……☐2

(b) If YES, does this limit your daily activity in any way?

Yes……☐1
No……☐2

THANK YOU FOR TAKING TIME TO COMPLETE THIS QUESTIONNAIRE ☺

9.25 STUDY 2 PCA SCREE PLOT OF EIGENVALUES FOR EACH COMPONENT NUMBER



9.26 STUDY 3 MATERIALS TRANSFER AGREEMENT (FIRST TWO PAGES)

MATERIAL TRANSFER AGREEMENT the "Agreement"

CONFIDENTIAL

LIVERPOOL JOHN MOORES UNIVERSITY

Agreement made on: 7th December 2016 (the Effective Date)

BETWEEN

1) **Liverpool John Moores University**
2) **Prof John Draper**
3) **Aberyswyth University**

Each a 'Party' and together the 'Parties'

DEFINITIONS

The Supplier:	Supplier of the Material being **Liverpool John Moores University whose principal office is at Egerton Court, 2 Rodney Street, Liverpool, L1 2UA**
The Recipient:	Individual to take receipt of the Material and use the Material in the Research programme being **Prof John Draper of Institute of Biological, Environmental and Rural Sciences**
The Institution:	The Recipient's Institution / employer being **ABERYSTWYTH UNIVERSITY**, (registered charity number 1145141), whose administrative offices are at Old College, King Street, Aberystwyth, Ceredigion SY23 2AX
The Material:	**1,200 x 4ml Urine Samples** This includes any constructs, strains, progeny, derivatives, portions, improvements and components (as the case may be) obtained from or as a result of the use of the Material.
Research Programme:	Description of research in which the Materials are to be used as set out in Schedule 1.
Intellectual Property:	all and any intellectual property rights of or relating to the Material, including but not limited to patents, trademarks, service marks, rights in a design and copyrights and any other intellectual property rights (whether or not registered or capable of registration) in any part of the world and the copyright and rights in the nature of copyright subsisting in all drawings, plans specifications, designs and computer software, know-how and Confidential Information.
Supplier Scientist	**Dr Leo Stevenson, Snr Lecturer Food Science, LJMU**
Term	**The term of this agreement being from the Effective date until 31st October 2018**

The Supplier has developed the Material. The Recipient is an employee of the Institution and wishes to acquire a sample of the Material for research relating to the Research Programme.

9.27 Study 3 Participant invitation (e-mail)

Dear Participant,

Thank you for completing the online 'Eating Habits Survey' as part of my PhD study on takeaway food consumers.

We have chosen you to take part in the next study and we would like to offer you an invitation but before you decide, please read the attached participant information sheet outlining the study in more detail. This study is to be carried out in the comfort of your own home at during a week that suits you.

If you decide you would like to take part in the study it would be greatly appreciated if you could reply saying that you are happy to take part. Then I will contact you and arrange the week that you would like to take part.

If you have any questions, please just send me an e-mail and I will get back to you as soon as possible.

Thank you very much for your help with this research.

Kind regards,

Hayley Janssen
Email – H.G.Janssen@2012.ljmu.ac.uk
Study mobile – 07970870622

9.28 Study 3 Participant information sheet (hardcopy)

Study Title: Determinants of Takeaway Food Consumption

By H. Janssen (Faculty of Education, Health and Community, LJMU)

Thank you for completing the online 'Eating Habits Survey'. We have chosen you to take part in the next study and we would like to offer you an invitation but before you decide, please read the details below.

1. **What is the purpose of the study?**

The aim of this study is to investigate patterns of takeaway food consumption among people in Merseyside. The aim of the collection of urine samples (in this study) is to identify novel chemicals (metabolite biomarkers) which are associated with takeaway/fast food diets/unhealthy eating.

2. **Do I have to take part?**

You are free to decide if you would like to take part or not and you can withdraw from the study at any time without giving a reason. To take part you should have **completed the 'Eating Habits Survey', be aged 18-64 years, eat takeaway/fast food** and **Live in Liverpool, Knowsley, St.Helens, Sefton, Wirral.**

3. **What will happen to me if I take part?**

This study is to be completed in the comfort of your own home during 1 week that suits you. We will ask you to complete three food records on three separate days (any days Mon-Sun) about the food you eat (you can eat anything you like). We will contact you by telephone to discuss your food records; this will take approximately 10 minutes each time. In addition, you will receive a urine collection-kit to provide two morning urine samples after each food record. The urine samples are stored in your fridge and once all six urine samples are collected you post them, along with the food records, in the pre-paid Safebox to any post box.

4. **What will happen to the samples collected?**

The urine samples will be stored at Liverpool John Moores University then sent to be examined at the laboratories in Aberystwyth University. All samples will be stored securely. We will perform tests to look for chemical markers of the different foods that you have eaten and to try to match to your eating pattern. After the study has finished the samples will be stored in our laboratory freezers in accordance with government regulations. Your name and details will no longer be associated with the samples.

5. **Are there any risks/benefits involved?**

No risks. After completing this part of the study you will be eligible to be entered into a prize draw to **win a £200 Love2Shop voucher**. We will ask you on the telephone if you are happy to be entered.

6. **Will my taking part in the study be kept confidential?**

Yes. Completion of the study is voluntary and anonymous; the information provided by you will be unidentifiable in any publications. Any personal information collected during this study will be stored securely on password protected computers or in locked filing cabinets.

7. **Contact Details of Researcher**

If you have any questions regarding your participation or the study please do not hesitate to contact:

H. Janssen, e-mail: **H.G.Janssen@2012.ljmu.ac.uk** Study mobile: **07970870622**

This study has received ethical approval from LJMU's Research Ethics Committee *(**16/EHC/002**). If you any concerns regarding your involvement in this research, please discuss these with the researcher in the first instance. If you wish to make a complaint, please contact researchethics@ljmu.ac.uk and your communication will be re-directed to an independent person as appropriate.*

9.29 Study 3 Written informed consent form

Study Title: Determinants of Takeaway Food Consumption

Researchers name - Hayley Janssen (Faculty of Education, Health and Community, LJMU)
Contact details: H.G.Janssen@2012.ljmu.ac.uk Study mobile: 07970870622

1. I confirm that I have read and that I understand the information provided for the above study. I have had the opportunity to consider the information, ask questions and have had these answered satisfactorily
2. I understand that my participation is voluntary and that I am free to withdraw at any time, without giving a reason and that this will not affect my legal rights
3. I understand that any personal information collected during the study will be anonymised and remain confidential
4. I consent to being contacted to take part in three food record interviews and provide six urine samples (two for each food record)
5. I consent for my urine samples to be stored in freezers at LJMU accessed only by authorised personnel
6. I consent for my urine samples to be sent to Aberystwyth University to be used for research purposes
7. I agree that samples collected will be stored after this study has completed and can be used for further research
8. I agree to take part in the above study

Name of Participant Date Signature

Name of Researcher Date Signature

Name of Person taking consent Date Signature
(if different from researcher)

Note: When completed 1 copy for participant and 1 copy for researcher
For office use only. Participant number: ………….

9.30 STUDY 3 URINE SAMPLING COLLECTION INSTRUCTIONS

Participant instructions – How to complete your diet record and collect urine samples

1

Check you have contents of kit: 6 test tubes, 6 absorbent pockets, sealable bag, 3 transfer straws, beaker, blue Safebox, 3 food records, 2 consent forms and blue pre-paid envelope.

2

Complete 2 consent forms; keep the white copy and place the blue copy in the blue pre-paid envelope. Keep the blue envelope for your food record and post once finished.

3

Complete food record of what you have eaten for 1 day. A urine sample will be collected the next morning. You will need to do this for 3 days (2 weekdays and 1 weekend day).

4

The morning after your food record, remove the red lid from the clear container without touching the inside of the container.

5

Fill the clear container at least three-quarters full with mid-stream urine.

6

Submerge the transfer straw well into the clear container but do not touch the bottom.

7

Insert one of the provided 4 mL sample tubes into the transfer straw. Make sure the test tube matches the food record; 1 (green), 2 (orange), or 3 (blue).

8

Whilst holding the transfer straw apply pressure down from the sample tube.

NOW REAPEAT STEPS 6-11

9

Keep pressure applied until sample tube is filled. The tube will not be filled completely.

10

Remove the transfer straw and sample tube from the urine filled clear container.

11

Remove the sample tube from the transfer straw and **fill identical test tube with the same urine sample.** Rinse the clear container with cold water and allow to air dry for reuse.

12

Please store samples in the fridge. Repeat this process for 3 days (2 weekdays, 1 weekend day) until all 3 food records and 6 samples are collected.

13

Use a new transfer straw for each identical set of samples. Keep all transfer straws, rinse with cold water and return in blue SafeBox as they contain a needle.

14

Place all sample tubes into the absorbent material pouches then roll up to fit in sealed plastic bag. **Store in fridge** until you are ready to send back.

15

Place the **6 samples** in the rolled up absorbent pouches in the sealable bag inside the blue Safebox along with the **3 transfer straws.** Seal as explained on the box.

16

Please post blue Safebox and blue pre-paid envelope on **Monday, Tuesday or Wednesday** to any post-box or post office near you.

Thank you ☺

Any questions please contact Hayley, e-mail: H.G.Janssen@2012.ljmu.ac.uk Study mobile: 07970870622

9.31 STUDY 3 FOOD RECORD FOR 24 HOUR 'MULTIPLE-PASS' DIETARY RECALL

Food record

Please answer the following questions:

1. Please enter today's date: ☐☐/☐☐/☐☐
   Day Month Year
2. Which day of the week is this record for? *Please tick one:*
   Sun ☐ Mon ☐ Tues ☐ Weds ☐ Thurs ☐ Fri ☐ Sat ☐
3. Is this a typical day? *Please tick one:* Yes ☐ No ☐ If No, Why?...............

Time & Place	**Details of Food and Drink (Name and Brand) *plus cooking method**	**Quantity eaten/Portion size**	**Food Checklist**
			• Sweets • Chocolate • Biscuits • Cakes • Crisps • Nuts • Pies/Pastry • Cheese • Milk • Spread • Fried Foods • Fruit • Vegetables • Pulses • Oily Fish • Red Meat
			Takeaway Food Checklist • Chinese • Indian • Kebab • Fried Chicken • Pizza
	Alcohol		

	Supplements/Medication (dose)		• Fish and Chips • Chips • Beef Burger
Has your diet changed since you completed the online FFQ? If so, how?			
Have you eaten any takeaway of fast food over the past 7 days? If YES, what days?			

9.32 STUDY 3 HUMAN TISSUE RISK ASSESSMENT

LIVERPOOL JOHN MOORES UNIVERSITY Life Sciences Building – COSHH Risk Assessment Form

Protocol/Procedure: Receipt of biological samples for PhD Study 16/EHC/002

Name of person carrying out procedure: Hayley Janssen	
Job Title: PhD Student	**Tel no. & e-mail:** 07922637667 H.G.Janssen@2012.ljmu.ac.uk
Faculty: EHC	**School:** Sport Studies, Leisure and Nutrition
Name of supervisor: Leo Stevenson **Tel no. & e-mail:** L.Stevenson@ljmu.ac.uk 01512315466 **Signature of supervisor:** Signed on page 4	
Location of procedure to be carried out: Room LSB1.25, 1st Floor, Life Sciences Building, Byrom Street, L3 3AF	
Is training/supervision required? No **Has the person been trained in this protocol/procedure?** No **If yes, please give details:**	
Date of assessment: 10/04/17 **Signature of person carrying out procedure:** **Signed on page 4**	

Description of Activity/Procedure/Process (include standard operating procedures (SOPs) as a Control): 1. Put on appropriate PPE (nitrile gloves and Howie laboratory coat); 2. Clear surfaces to ensure enough space to work; 3. Open SafeBox and check carefully for signs of leakage; 4. If there is no evidence of leakage, put the SafeBox into the general waste (black bin); 5. Discard of the sharps in a sharps-bin 6. Remove samples from inner packet and absorbent packaging, place packaging into clinical waste (yellow) bin; 7. Put sample into the freezer. 8. Remove gloves and wash hands
What are the potential risks?

Participants of this study are presumed to be healthy and have no known infectious conditions. However, biological samples may contain a variety of infectious agents (viruses, bacteria etc.) Even ostensibly clean containers may contain residual biological matter. This potentially could cause infection. Sharps could pierce the skin and gloves, causing needle-stick injury. This is an increased route for infection.
What personal protective equipment (PPE) is required? (ref:SCP9) Nitrile gloves and Howie Lab coat.
What else can be done to mitigate the risks? • Safe Boxes and vacutainers are being used to reduce the likelihood of breakage during transport. • Boxes will be opened carefully, in the laboratory environment. • SafeBoxes will be frozen upon receipt in Goods Reception (G35) and therefore any liquid that has leaked within the packaging should be solid upon opening in the LSB. • Participants have been asked to place the 4ml vacutainers into absorbent packaging, within a sealed bag to reduce any leakage. • Sharps will be placed carefully into a clinical waste sharps-bin
Who is at risk? (staff/students/others): Anyone else using the room.
What is the duration of exposure? 1-2 hours
What is the frequency of exposure? Weekly
What is the maximum number of people in the room/lab: 3
Are there any ethical issues: Yes **If yes, has consent been obtained?** Yes
Do any additional personal/medical risks apply? (e.g. pregnancy/asthma etc.); if so discuss this with your Supervisor/Senior Research Officer before completing the form.
Health Surveillance is required if the procedure involves substances which are respiratory or skin sensitizers: H317 (may cause an allergic skin reaction) or H334 (may cause allergy or asthma symptoms or breathing difficulties if inhaled). If the chemicals listed below have hazard codes: H317 or H334 then consult with Occupational Health Unit (OHAdmin@ljmu.ac.uk) to discuss the requirement for health surveillance. **Do the chemicals listed below have the hazard codes H317/H334?** No **Is health surveillance required (if no, state why)?** N/A If other substances with potential health effects are used and if any health effects are observed that is believed to have resulted from its use then Occupational Health Unit should be contacted. Consideration should be made of the existing health status of the user of hazardous substances (e.g. An asthmatic working with respiratory irritants). **Are special arrangements required?** (e.g. for additional types of PPE): No
Will the individual be working outside of normal working hours, overnight or at weekends? No **If yes, has the appropriate form (SCP11 – Out of hours Working) been completed and authorisation given?** N/A **Will the experiment be left to run unattended out of hours?** N/A **If yes, has the appropriate form (SCP22 – Unattended Experiments) been completed and authorisation given?** N/A

Starting substances:	Urine
How will these risks be controlled?	PPE is worn to protect areas exposed such as skin & sharps bin.
End products:	Same as above
Hazard categories:	N/A

Transfer/Storage/Disposal/Spillage.

Chemical storage: N/A
Transfer procedure: Samples will be transported from Goods Reception (G35) to LSB 1.25 in unopened Safe-Boxes.
Waste disposal procedure: Potentially contaminated packaging and waste into clinical waste for incineration.

Spillage/leaks procedure:
Two types of leakage/spillage may occur:
i). Confined to the inner bag and absorbent packaging ii) Breached the inner bag and contaminating the safe box and potentially laboratory benches.

Procedure:
i). The bag should be placed in a 1L beaker containing 500ml of 2% Virkon and the bag opened to allow sufficient mixing with the disinfectant. After 30 minutes any liquid waste can be disposed down the laboratory sink. Any solid waste can go into a clinical waste bin.
ii). Any loose sample (solid/liquid) should be decanted into a beaker containing 500ml 2% Virkon and left for at least 30 minutes. A spray bottle containing 2% Virkon should be used to spray the inside of the container and left for 30 minutes. Any liquid waste can be disposed down the laboratory sink, solid waste can go into the clinical waste bin.

In the event of a large spillage or accidental release, the following procedures should be followed:

- Apply Virkon powder (under the sink in LSB 1.26) directly to the spill. Leave for 5 minutes. Mop-up using lab roll and dispose in the clinical waste bin.

In case of fire or explosion:

Raise alarm, immediately evacuate area, call security on ext. 2222 (0151 231 2222 from an external line) and inform them of the location and source of the fire, they will contact the fire brigade. Inform fire marshal on site of the location and source of the fire.

In case of emergency please contact:

Please add names and contact details (telephone number required) for your supervisor:

Supervisor(s):

- Local First Aiders: a list can be found on the LJMU Health and Safety Unit webpage https://www2.ljmu.ac.uk/HSU/index.htm. Please check that these are up-to-date.

Joe Furmage	Ground Floor	Ext. 2218
Kenny Ritchie	Room 1.13	Ext. 2219
Jerry Bird	Room 2.06	Ext. 2181

- Local Health and Safety Officers: a list can be found on the LJMU Health and Safety Unit webpage https://www2.ljmu.ac.uk/HSU/index.htm. Please check that these are up-to-date.

School	H&S Officer	Email	Telephone
SPS	Monica Barclay	m.barclay@ljmu.ac.uk	TRB Ext. 6211 (0151 904 6211)
PBS	Kathryn Farrell	k.l.farrell@ljmu.ac.uk	JP Ext. 2315 (0151 231 2315)
NSP	Dr Jerry Bird	J.M.Bird@ljmu.ac.uk	LSB Ext 2181 (0151 231 2181)

In case of business interruption (e.g. power shutdown, flood), what are the Contingency Procedures for work and waste?

In the event of a power cut the Cryo-storage vessel should be left with the lid firmly in place until work can resume. The room should be vacated until oxygen level alarms are operational.

Emergency action procedures – action required if a substance is:

Exposure	Action
Inhaled	Not considered a potential route of exposure.
Ingested	Not considered a potential route of exposure
Contact with skin	Wash thoroughly with soap and water.
Contact with eye	Rinse immediately with plenty of water, also under the eyelids, for at least 15 minutes. Seek medical advice.
Injected	Injection is not considered a potential route of exposure.

Based on the COSHH information and the safety measures put in place to minimise risk, is the overall risk of your procedure/protocol high, medium or low? Low

For further information contact Health and Safety Unit (3214/3375), refer to supplier's data sheet, contact supervisor.

9.33 Study 3 Goods reception protocol

<u>Goods G35 protocol for dark blue SafeBoxes (urine samples) and pale blue envelopes (consent forms) for PhD study</u>

SafeBoxes (fig. 1) - please store in freezer as soon as they arrive at Goods Reception G35
Blue envelopes (fig. 2) please store in safe place

↓

Contact Hayley on arrival:
H.G.Janssen@2012ljmu.ac.uk
Work mobile: 07970 870622
I will usually collect Safeboxes and blue envelopes on Tuesdays or Wednesdays and transfer them to freezers in Room LSB1.25, 1st Floor, Life Sciences Building

↓

Figure 1. Safebox – urine

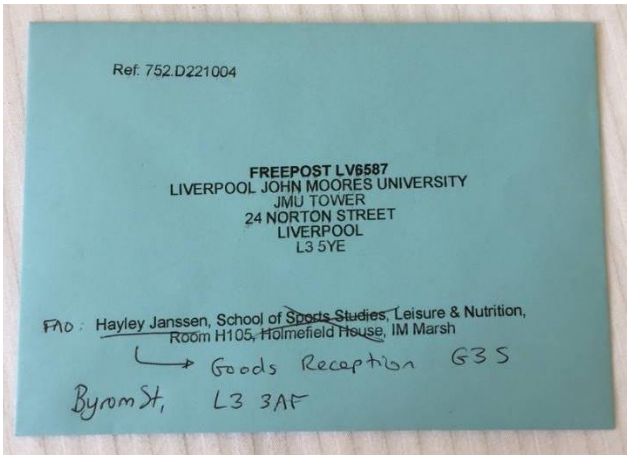


Figure 2. Blue envelope – consent form

If freezer space is unavailable and Hayley cannot be reached, please contact:
John Hall: J.S.Hall@ljmu.ac.uk 01512312431
OR
Alan Simm: A.M.Simm@ljmu.ac.uk 01512312214

9.34 STUDY 3 ACCEPTABILITY QUESTIONNAIRE INVITATION

Dear Participant,

Thank you for completing the urine collection study.

We would now like to invite you to complete a very short questionnaire (13 tick boxes and 4 questions) regarding the acceptability of the home urine sampling kit. This will take approximately **2 minutes** to complete. Before you decide please read the participant information sheet attached.

We would really appreciate your feedback, as this is a completely new method that we hope to improve based on your experience.

Acceptability questionnaire;
https://ljmu.onlinesurveys.ac.uk/acceptability-questionnaire

Many thanks,
Hayley

Hayley Janssen, MPhil, BSc (Hons), AFHEA, SEDA, ANutr.
PhD Researcher
Food, Nutrition and Health Research Group
Faculty of Education, Health & Community
IM Marsh Campus, Barkhill Road, Liverpool, L17 6BD
e: H.G.Janssen@2012.ljmu.ac.uk

9.35 STUDY 3 ACCEPTABILITY QUESTIONNAIRE PARTICIPANT INFORMATION SHEET

Study Title: Determinants of Takeaway Food Consumption
By H. Janssen (Faculty of Education, Health and Community, LJMU)

Thank you for completing the urine collection study. We would now like you to provide some feedback regarding the bespoke urine sampling collection-kit but before you decide, please read the details below.

1. **What is the purpose of the study?**

The aim of this part of the study is to find out your opinion of using this new method of collecting urine in a home setting and how acceptable you found it in practice.

2. **Do I have to take part?**

You are free to decide if you would like to take part or not and you can withdraw from the study at any time without giving a reason. To take part you should have **completed the urine collection study.**

3. **What will happen to me if I take part?**

The 'Acceptability Survey' takes approximately 2 minutes to complete. The questions are asked in a way to rate your opinion towards a statement concerning the method on a scale from strong agreement to strong disagreement with the statement. There is also an opportunity to add any extra comments regarding the method.

4. **Are there any risks/benefits involved?**

No risks.

5. **Will my taking part in the study be kept confidential?**

Yes. Completion of the study is voluntary and anonymous; the information provided by you will be unidentifiable in any publications. Any personal information collected during this study will be stored securely on password protected computers or in locked filing cabinets.

6. **Contact Details of Researcher**

If you have any questions regarding your participation or the study please do not hesitate to contact:
H. Janssen, e-mail: **H.G.Janssen@2012.ljmu.ac.uk** Study mobile: **07970870622**

This study has received ethical approval from LJMU's Research Ethics Committee (*16/EHC/002*). *If you any concerns regarding your involvement in this research, please discuss these with the researcher in the first instance. If you wish to make a complaint, please contact researchethics@ljmu.ac.uk and your communication will be re-directed to an independent person as appropriate.*

Many thanks! ☺

9.36 STUDY 3 ACCEPTABILITY QUESTIONNAIRE

Questionnaire on the acceptability of home urine collection and storage

Welcome

Thank you for agreeing to take part in this survey and urine collection study. By completing this survey, you are consenting to be part of this research study. All information will be treated in the strictest confidence.

Thank you for your time.

Contact details of researcher

If you have any questions about the study or about taking part please contact: Hayley Janssen, e-mail: **H.G.Janssen@2012.ljmu.ac.uk**
This study has received ethical approval from LJMU's Research Ethics Committee *(16/EHC/002)*. *If you any concerns regarding your involvement in this research, please discuss these with the researcher in the first instance. If you wish to make a complaint, please contact researchethics@ljmu.ac.uk and your communication will be re-directed to an independent person as appropriate.*

Instructions

We are interested in finding out your opinion of using this new method of collecting urine in a home setting and how acceptable you found it in practice.

The questions are asked in a way to rate your opinion towards a statement concerning the method on a scale from **strong agreement** to **strong disagreement** with the statement. Please estimate your opinion to the stated as best as you can and fill in the circle that is closest to match with it. Fill-in only one circle for each question.

Example:

Would it be difficult to follow the

○	○	●	○	○
Strongly disagree	Disagree	Neither agree nor disagree	Agree	Strongly agree

For example, if you fill in this circle, you are saying that it is

Please read these statements carefully. Fill in the circle that is the closest match to your opinion where the **leftmost is not at all agreeing with the statement** and the **rightmost is strongly agreeing with the statement**.

1. I was successful in collecting urine using this method ○ Strongly disagree ○ Disagree ○ Neither agree nor disagree ○ Agree ○ Strongly agree
2. It was easy to collect urine in the pot ○ Strongly disagree ○ Disagree ○ Neither agree nor disagree ○ Agree ○ Strongly agree
3. I was confident collecting urine in the pot ○ Strongly disagree ○ Disagree ○ Neither agree nor disagree ○ Agree ○ Strongly agree
4. It was easy to transfer urine from the pot into the capped tube using the straw ○ Strongly disagree ○ Disagree ○ Neither agree nor disagree ○ Agree ○ Strongly agree
5. I felt confident transferring urine from the pot into the capped tube using the straw ○ Strongly disagree ○ Disagree ○ Neither agree nor disagree ○ Agree ○ Strongly agree
6. I would be happy to write the collection date and time on the ○ Strongly disagree ○ Disagree ○ Neither agree nor disagree ○ Agree ○ Strongly agree **capped tube**
7. I was happy collecting first morning void samples ○ Strongly disagree ○ Disagree ○ Neither agree nor disagree ○ Agree ○ Strongly agree

8. I would have preferred to collect urine samples at a different time of day

○ Strongly disagree	○ Disagree	○ Neither agree nor disagree	○ Agree	○ Strongly agree

9. I was happy storing several urine samples collected over a week in my fridge

○ Strongly disagree	○ Disagree	○ Neither agree nor disagree	○ Agree	○ Strongly agree

10. I was happy to post urine samples in a pre-paid box

○ Strongly disagree	○ Disagree	○ Neither agree nor disagree	○ Agree	○ Strongly agree

General questions

1. In general, I think collecting an urine sample is difficult

○ Strongly disagree	○ Disagree	○ Neither agree nor disagree	○ Agree	○ Strongly agree

2. I think collecting an urine sample in a home environment is embarrassing

○ Strongly disagree	○ Disagree	○ Neither agree nor disagree	○ Agree	○ Strongly agree

3. I think collecting an urine sample OUT of the home environment is embarrassing

○ Strongly disagree	○ Disagree	○ Neither agree nor disagree	○ Agree	○ Strongly agree

Any additional comments regarding the urine collection-kit?

When were the urine samples posted in the safebox? *Please select one answer*

- The last day of urine sampling
- 1-2 days after
- 3-4 days after
- 4-7 days after
- Over a week after

Has your diet changed since completing the online 'Eating Habits Survey? For example, joined a weight loss group or changed to a vegetarian/vegan diet

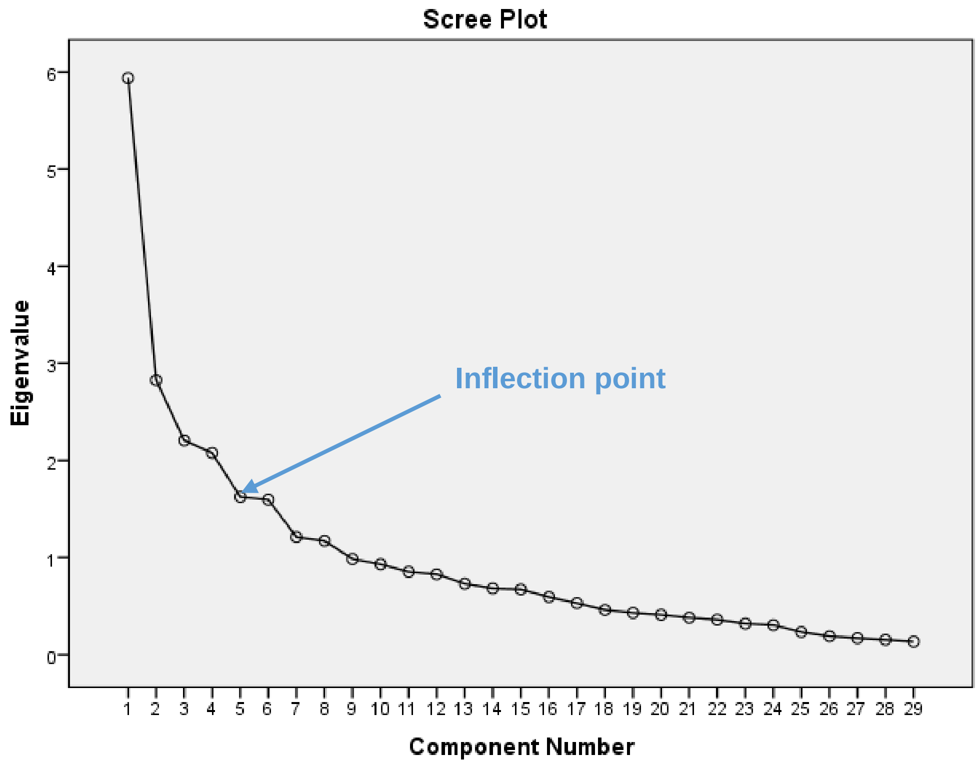
- No
- Yes

If yes, please tell us in what way has your diet changed?

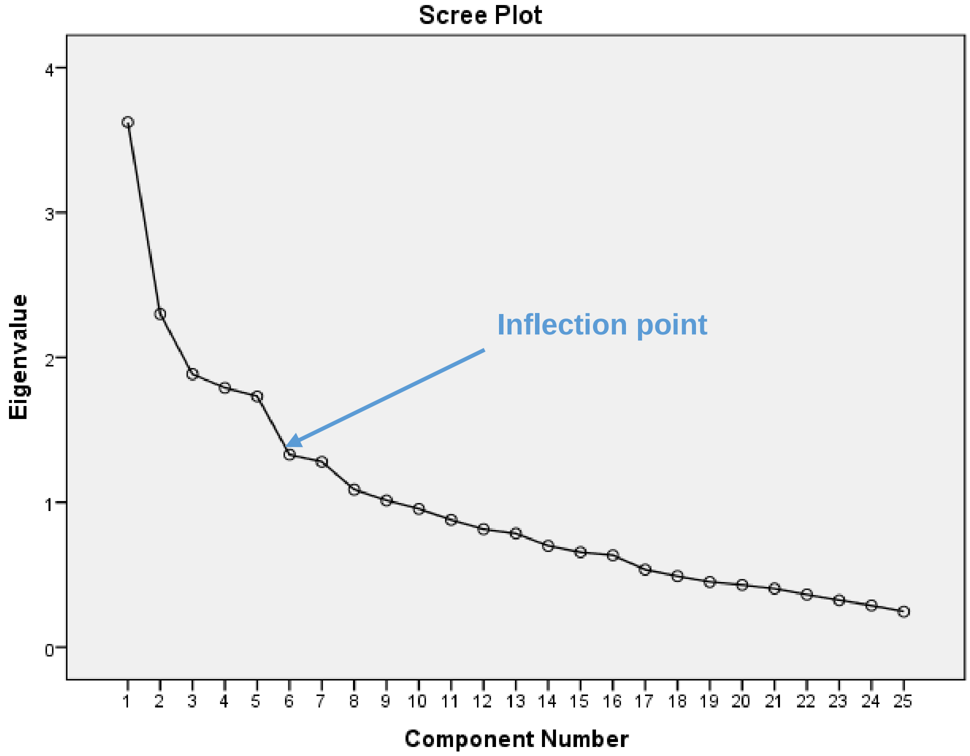
Lastly, to be able to correlate your responses with the samples taken and your demographics please can we have your name and email?

Name: Email address:

9.37 STUDY 3 PCA SCREE PLOT FROM FFQ OF EIGENVALUES FOR EACH COMPONENT



9.38 STUDY 3 PCA SCREE PLOT FROM 24 H MPR OF EIGENVALUES FOR EACH COMPONENT



9.39 STUDY 3 PCA COMPONENT MATRIX (UN-ROTATED) 24 H MPR

Component Matrix[a]

	Component				
	1	2	3	4	5
Vegetables	-0.597	0.247	0.288	0.271	0.184
Other bread, pasta, cereal, rice	0.590	0.430	0.148	0.224	0.113
Nuts and seeds	-0.562	0.145	-0.150	0.263	0.248
Wholegrain bread, pasta, cereal, rice	-0.562	0.093	0.115	0.233	-0.266
Fruit	-0.530	0.169	0.256	0.071	-0.128
Carbonated soft drink or cordial	0.506	0.110	-0.314	0.360	-0.099
Legumes (including soy)	-0.482	0.187	-0.178	0.364	0.475
Fast Food	0.470	-0.326	-0.176	0.408	-0.159
Potato fried (chips, roasties, fries)	0.436	-0.285	-0.001	0.428	-0.137
Red and processed meat	0.422	-0.010	0.226	-0.218	0.000
High Salt Snacks	0.355	0.165	-0.131	-0.085	0.350
Sugar inc honey	0.332	0.279	-0.141	-0.193	0.238
Dairy	0.194	0.715	0.127	-0.116	-0.303
Buns, cakes, pastries and fruit pies	0.390	0.554	-0.042	0.132	-0.068
Coffee	-0.210	0.484	0.201	0.023	-0.381
Pizza takeaway	0.240	0.443	-0.023	0.227	-0.067
Cocoa	0.253	0.419	-0.050	-0.254	0.133
Poultry (chicken and turkey)	0.232	-0.357	0.151	0.232	-0.277
Sugary condiments (chutneys, HP sauce)	0.159	-0.132	0.715	0.152	0.282
Carbonated soft drink DIET/Sweeteners	0.142	-0.080	0.550	-0.168	0.382
Chinese takeaway	0.256	-0.225	0.343	0.194	-0.034
Oils and dairy free spreads	0.237	0.121	0.346	0.572	0.071
Fruit Juice	-0.180	0.106	-0.158	0.399	0.333
Egg	-0.204	0.068	0.410	-0.066	-0.493
Biscuits	0.205	-0.034	0.317	-0.231	0.390

NB: Major positive loadings for each item are in red (factor loadings > 0.3 are considered significant). Extraction method: Principal component analysis. Rotation method: Un-rotated

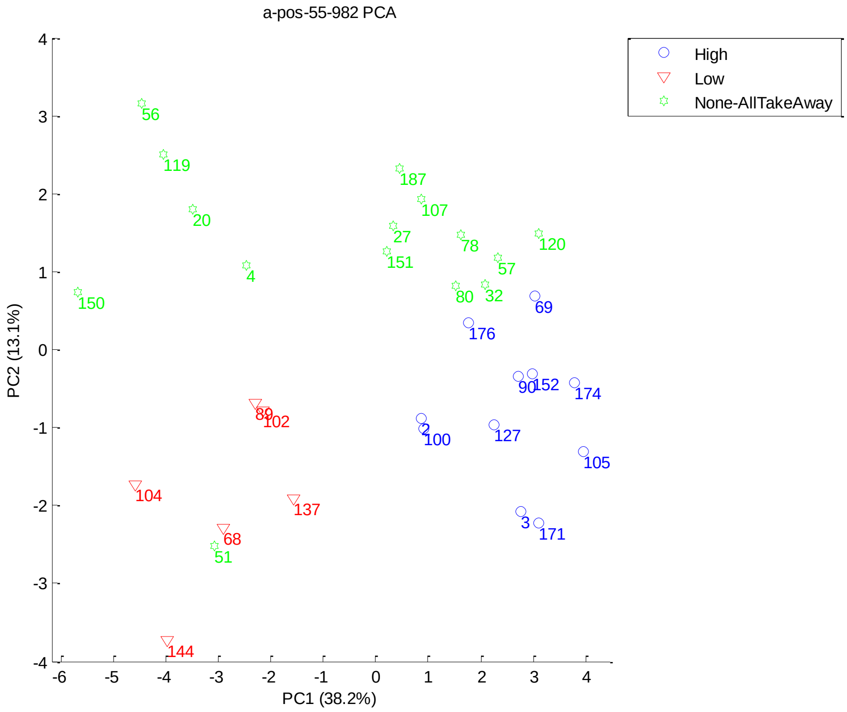
9.40 Study 3 PCA Component Matrix (un-rotated) matrix FFQ

Component Matrix[a]

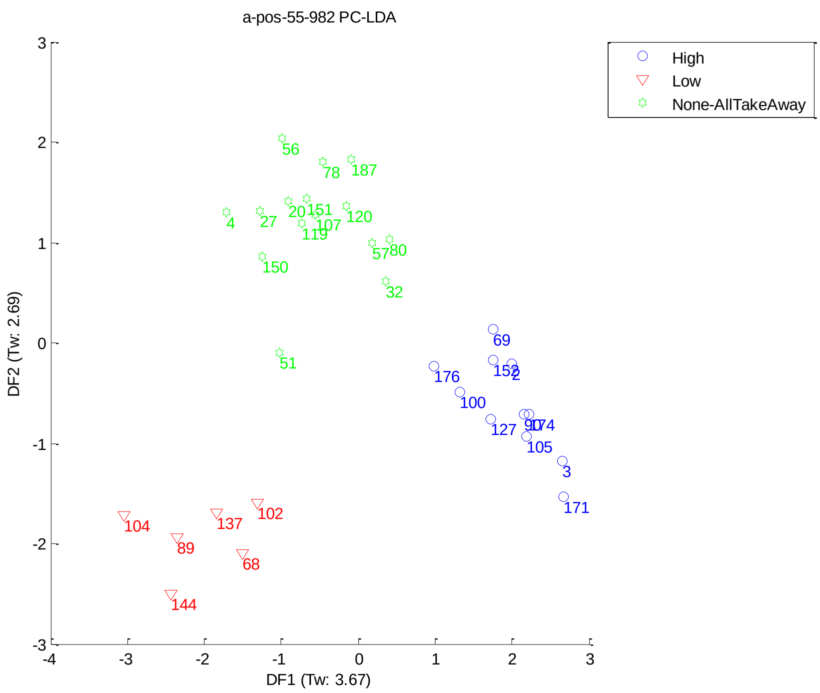
	Component			
	1	2	3	4
Cakes Pastry Buns	0.752	0.207	-0.263	-0.161
Fried Potato	0.730	-0.063	0.070	0.098
Other Grains	0.708	0.050	-0.306	0.208
Other Potato	0.696	0.116	0.048	0.010
High Salt Snacks	0.574	-0.123	-0.228	-0.071
Indian	0.547	0.195	0.127	0.392
Legumes	0.540	-0.022	0.507	-0.383
English	0.534	-0.463	0.378	-0.011
Sweet Sauces	0.490	-0.345	0.338	-0.296
Sweets and Sugar	0.485	-0.265	-0.132	0.093
SSB	0.479	-0.266	0.236	0.060
Biscuits	0.431	-0.205	-0.361	-0.326
Savoury Sauces	0.426	0.311	0.182	0.039
Vegetables	0.378	0.623	0.304	-0.019
Fast Food	0.504	-0.537	0.069	0.115
Fruit	0.023	0.531	0.262	0.049
Wholegrains	0.172	0.504	0.196	-0.079
Nuts	0.363	0.466	0.247	-0.230
Fish	0.256	0.461	0.103	0.271
Diet SB	0.185	-0.252	0.087	-0.192
Dairy	0.493	0.330	-0.543	-0.122
Eggs	0.155	0.280	-0.532	-0.180
Fats and Oils	0.392	0.188	-0.462	-0.130
Pizza	0.126	0.140	0.348	-0.174
Kebab	0.178	-0.150	-0.062	0.618
Chinese	0.493	-0.096	0.034	0.505
Red Meat	0.244	-0.108	-0.060	0.471
Chocolate	0.379	-0.326	-0.119	-0.425
Poultry	-0.025	0.010	0.010	0.406

NB: Major positive loadings for each item are in red (factor loadings > 0.3 are considered significant). Extraction method: Principal component analysis. Rotation method: Un-rotated

9.41 STUDY 3 METABOLITE FINGERPRINTS FOR INDIAN TAKEAWAY

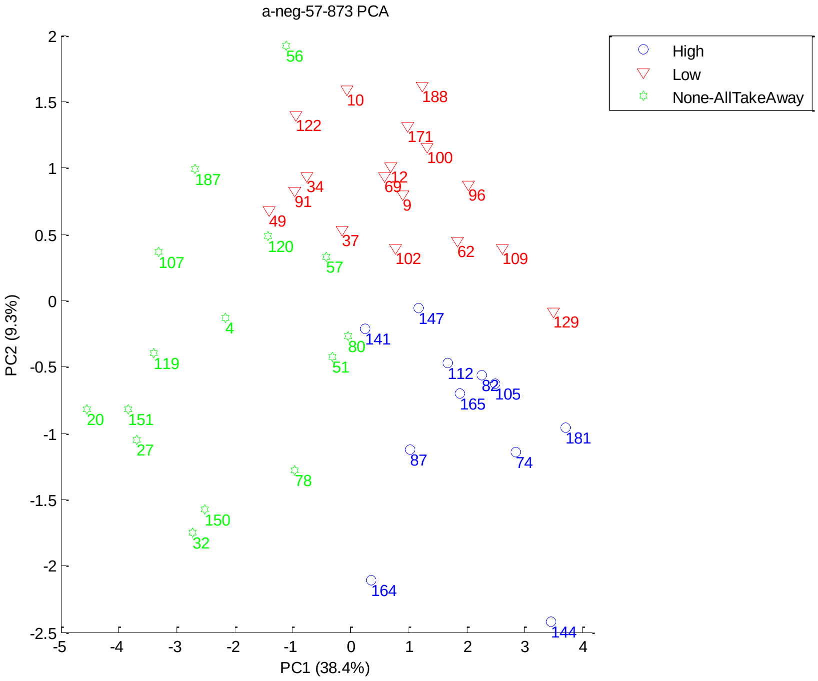


PCA plot Positive mode m/z 55 – 982 for Indian takeaway

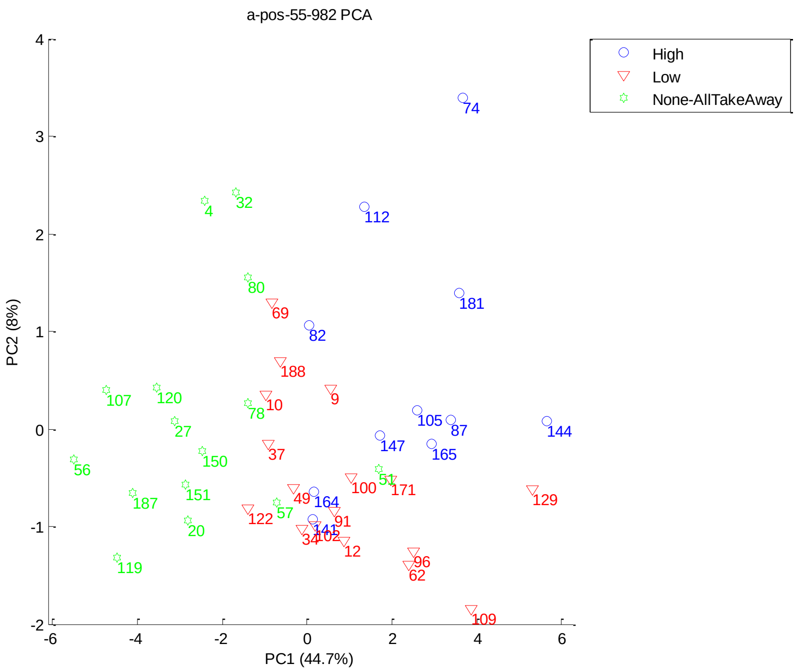


PC-LDA plot Positive mode m/z 55 - 982 for Indian takeaway

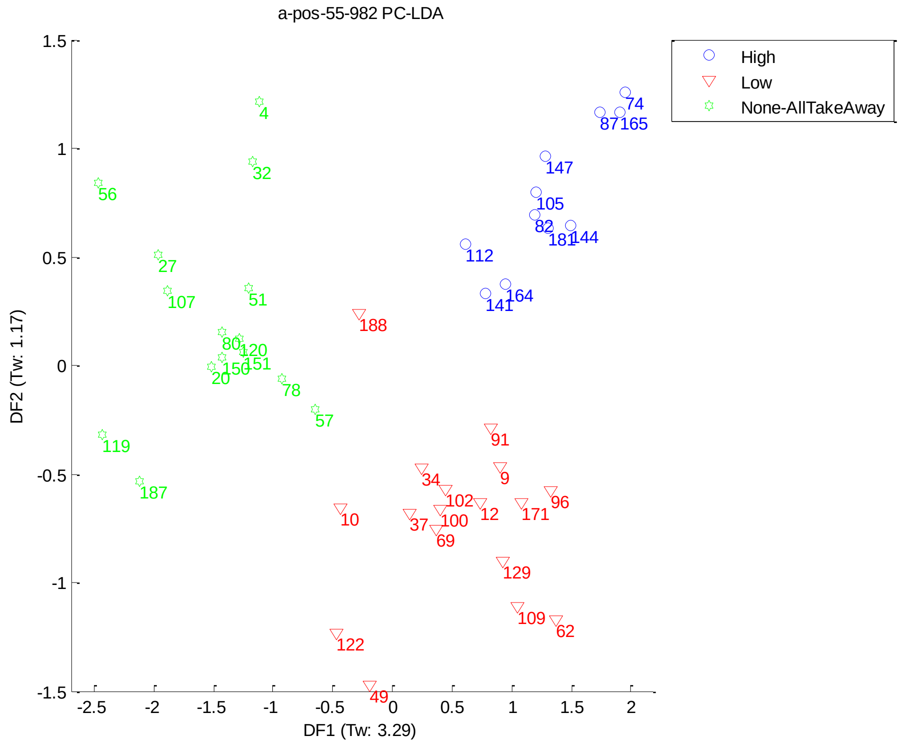
9.42 STUDY 3 METABOLITE FINGERPRINTS FOR FAST FOOD



PCA plot Negative mode m/z 57-873 for Fast Food

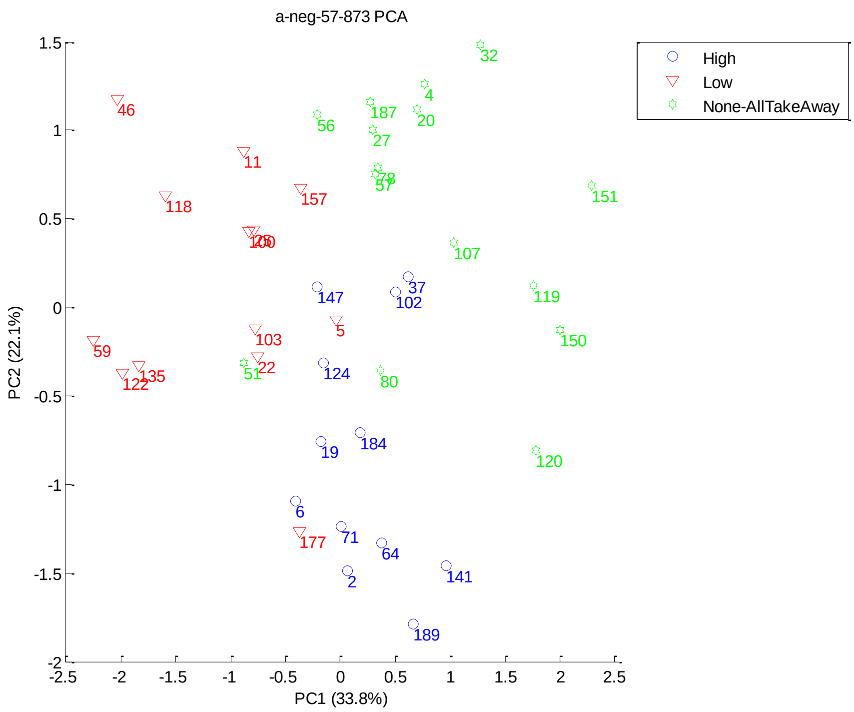


PCA plot Positive mode m/z 55 – 982 for Fast Food

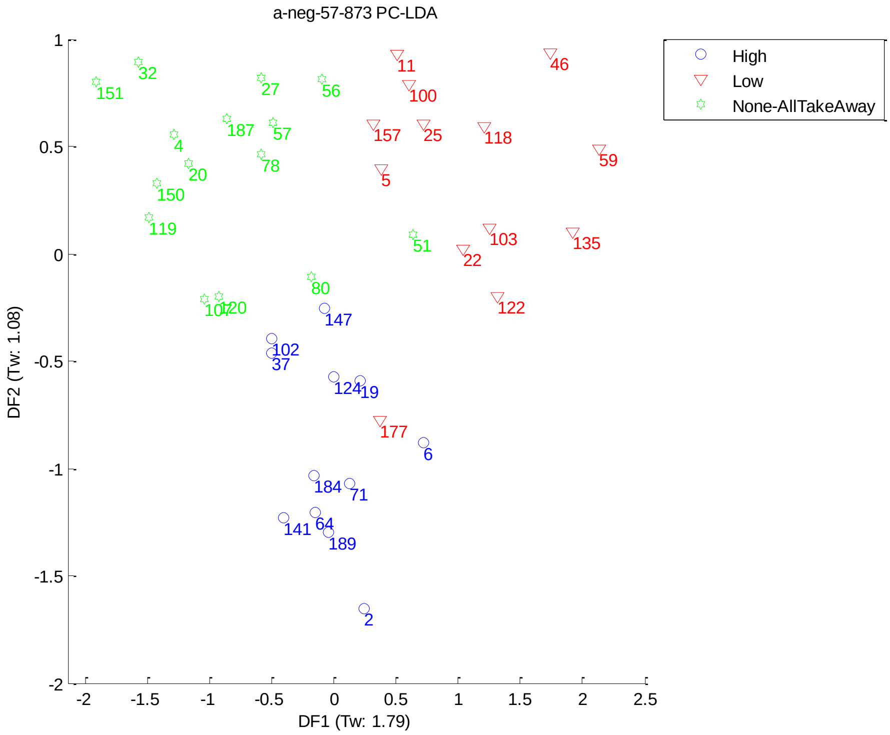


PC-LDA plot Positive mode m/z 55 – 982 for Fast Food

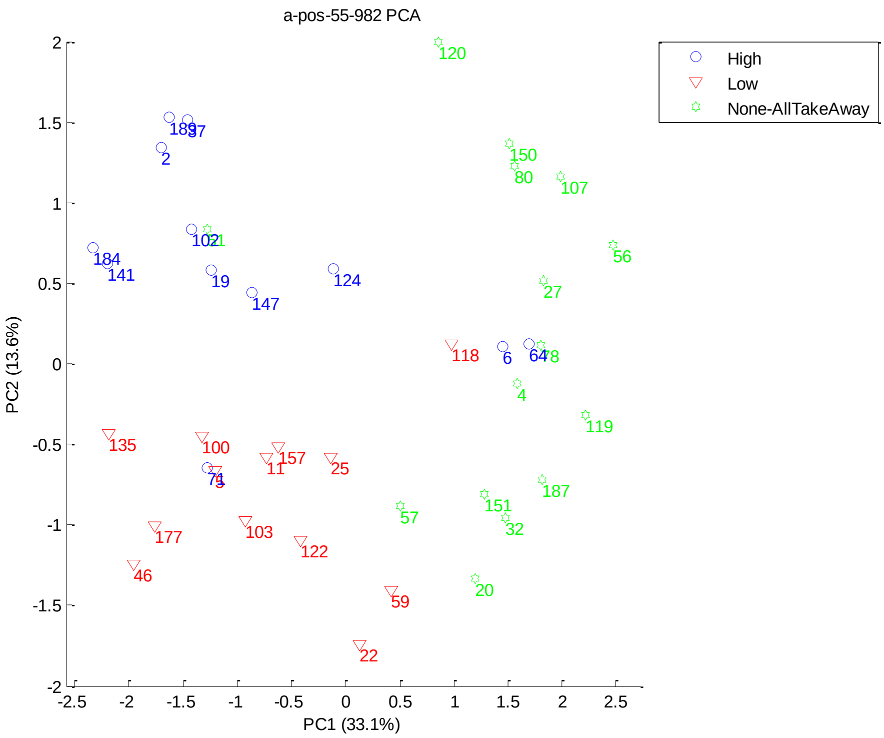
9.43 Study 3 Metabolite fingerprints for Chinese takeaway



PCA plot Negative mode m/z 57-873 for Chinese

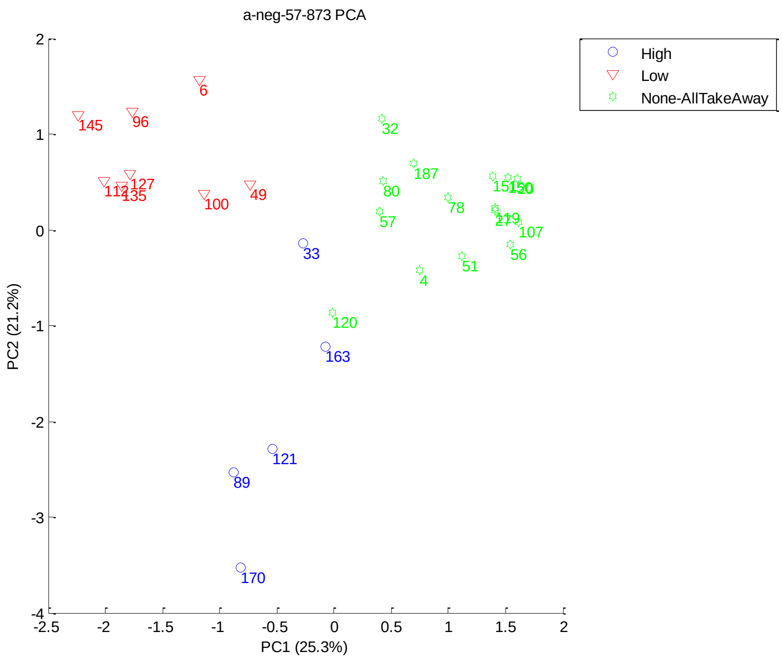


PC-LDA plot Negative mode m/z 57-873 for Chinese takeaway

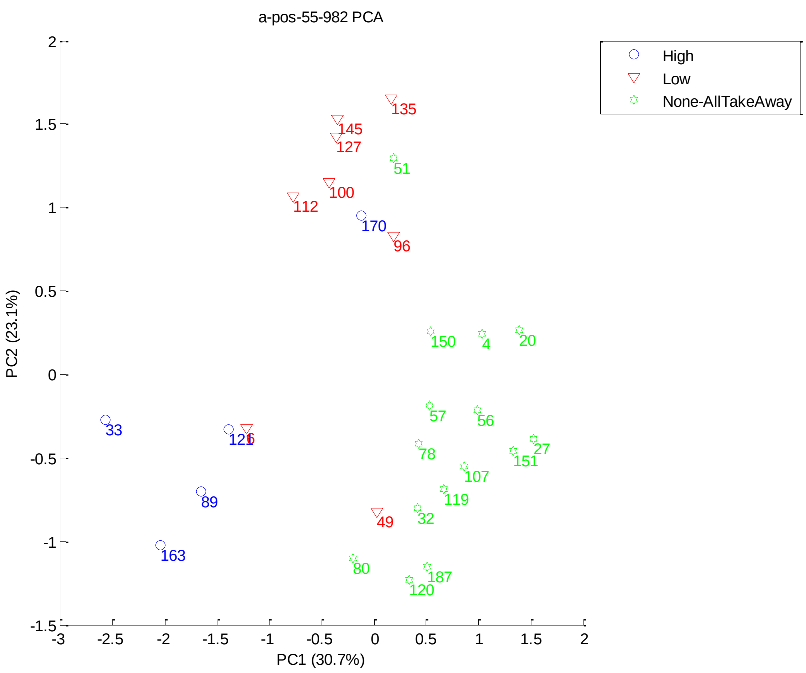


PCA plot Positive mode m/z 55 - 982 for Chinese takeaway

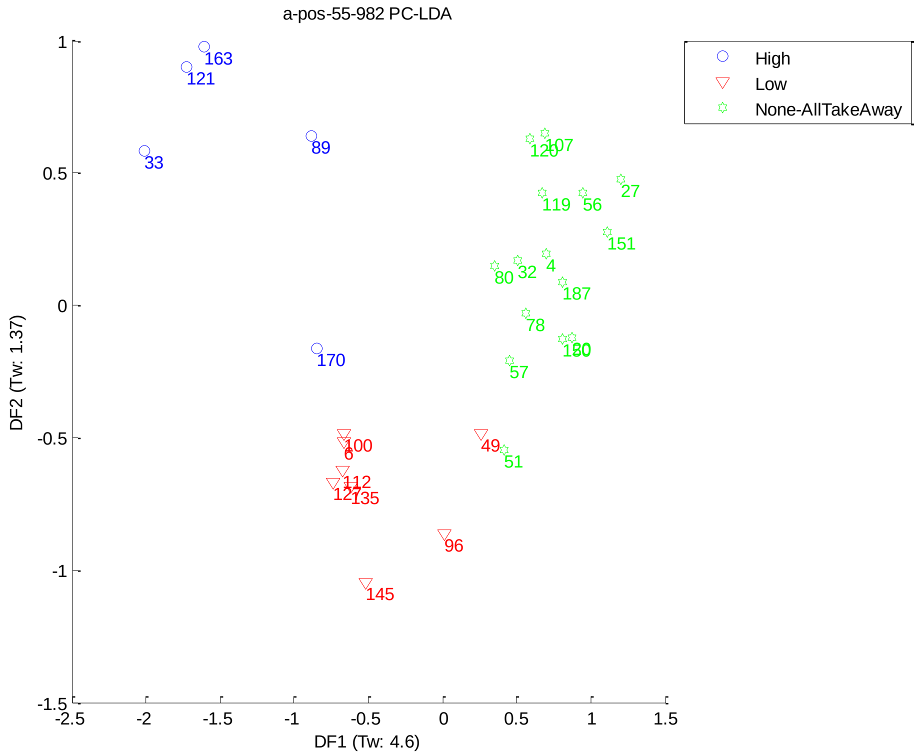
9.44 Study 3 Metabolite Fingerprints for English Takeaway



PCA plot Negative mode m/z 57-873 for English Takeaway

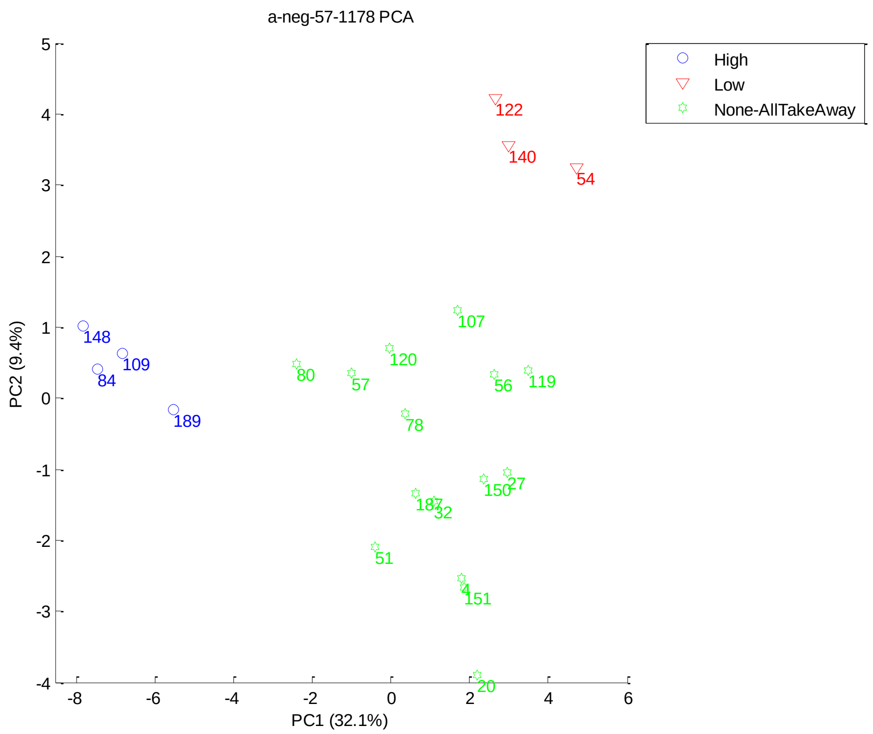


PCA plot Positive mode m/z 55 - 982 for English Takeaway

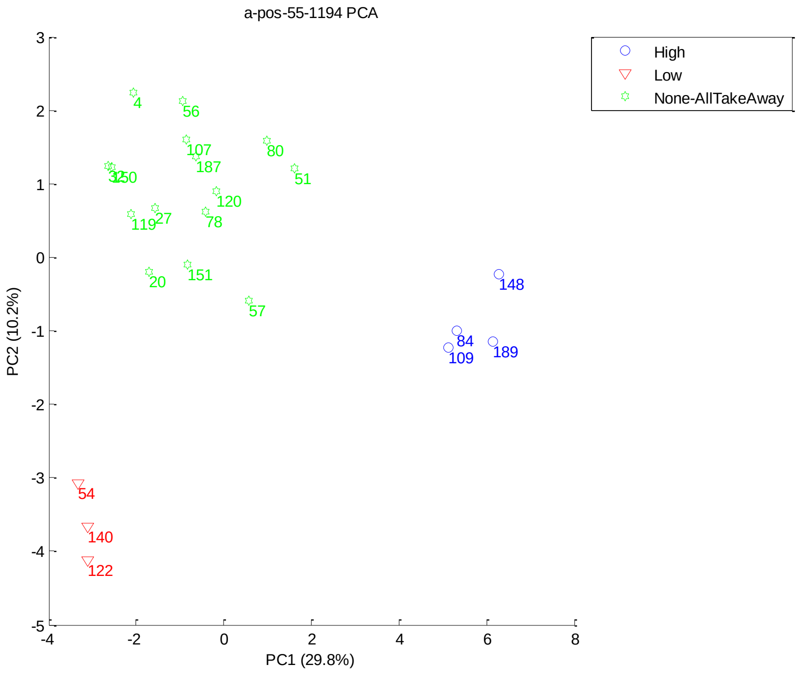


PC-LDA plot Positive mode m/z 55 - 982 for English Takeaway

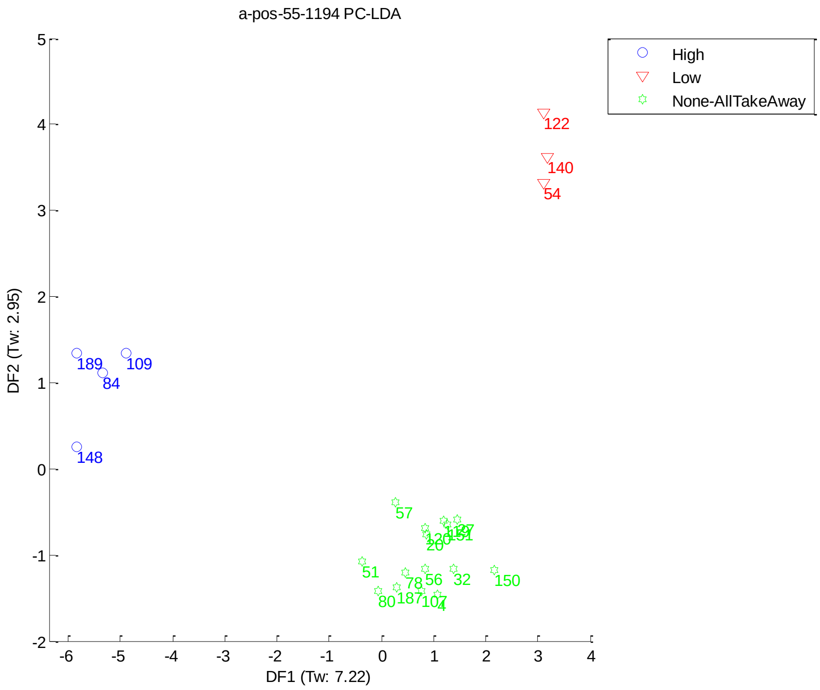
9.45 STUDY 3 METABOLITE FINGERPRINTS FOR KEBAB TAKEAWAY



PCA plot Negative mode m/z 57 - 873 for kebab

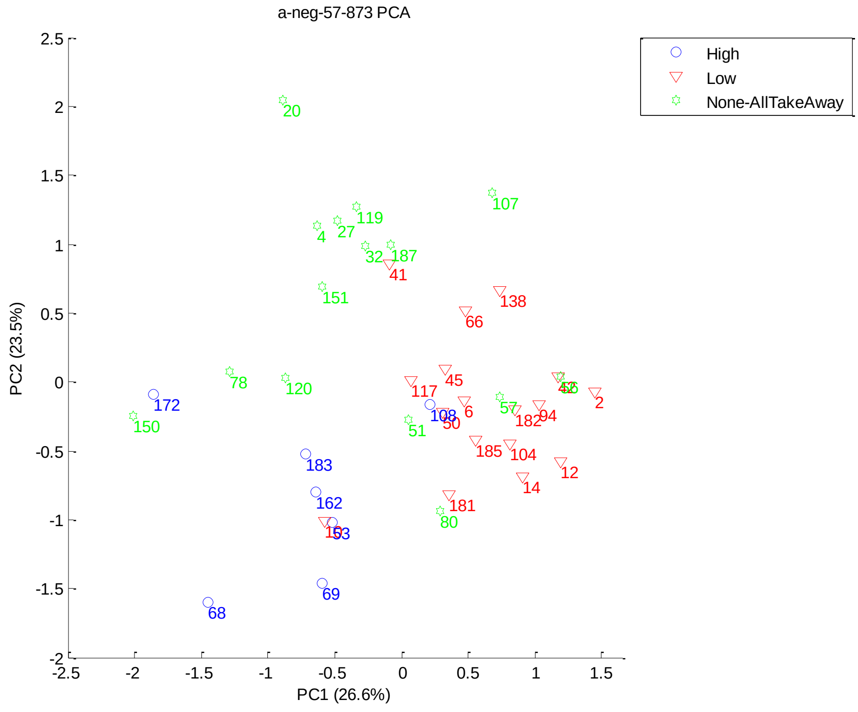


PCA plot Positive mode m/z 55 - 982 for kebab

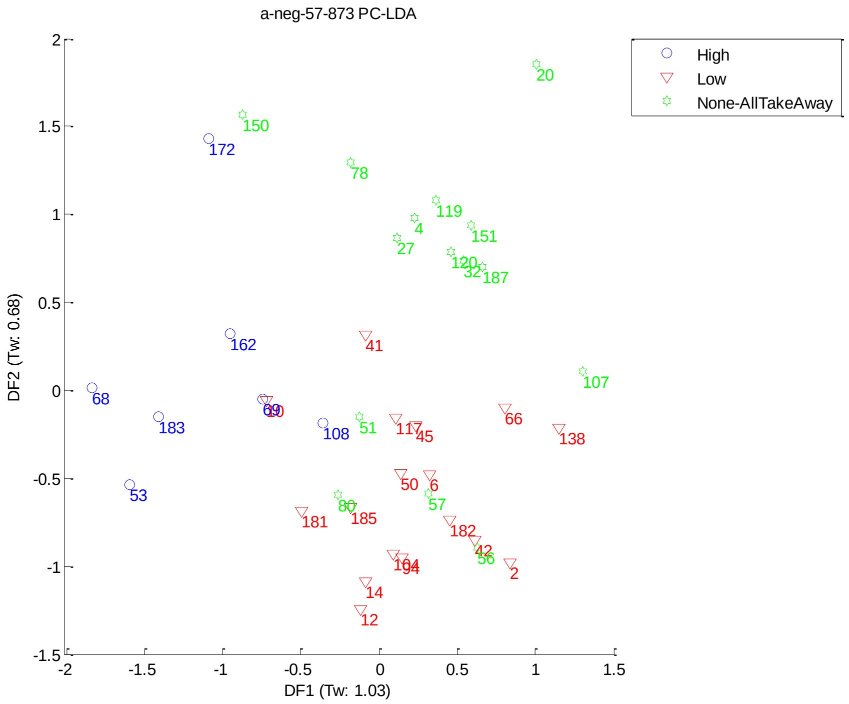


PC-LDA plot Positive mode m/z 55 - 982 for kebab

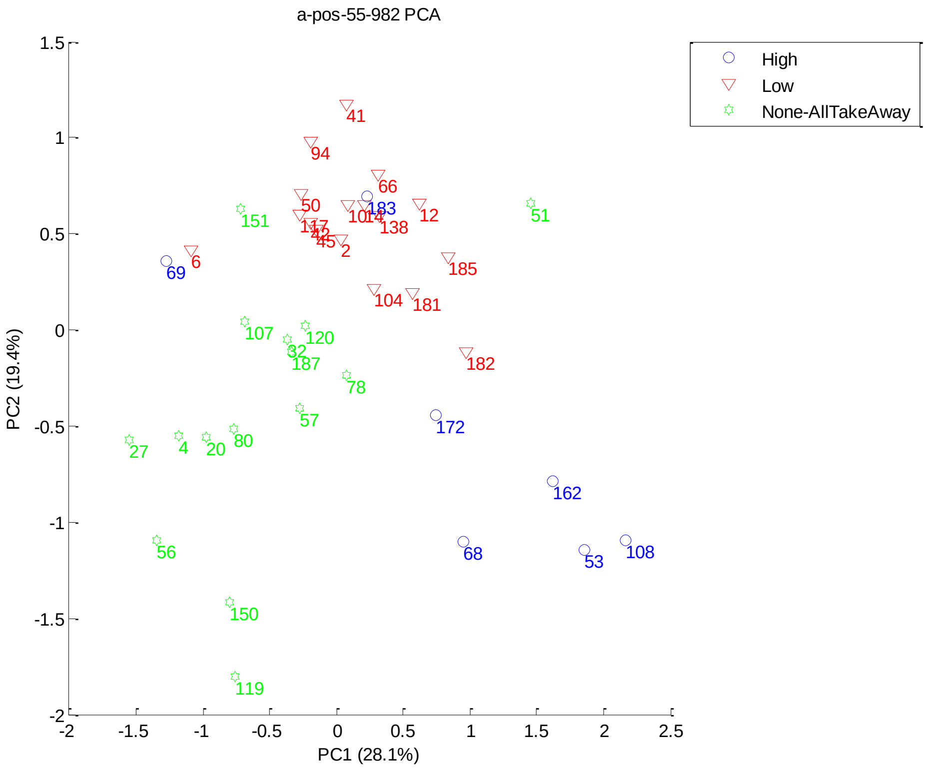
9.46 Study 3 Metabolite fingerprints for pizza takeaway



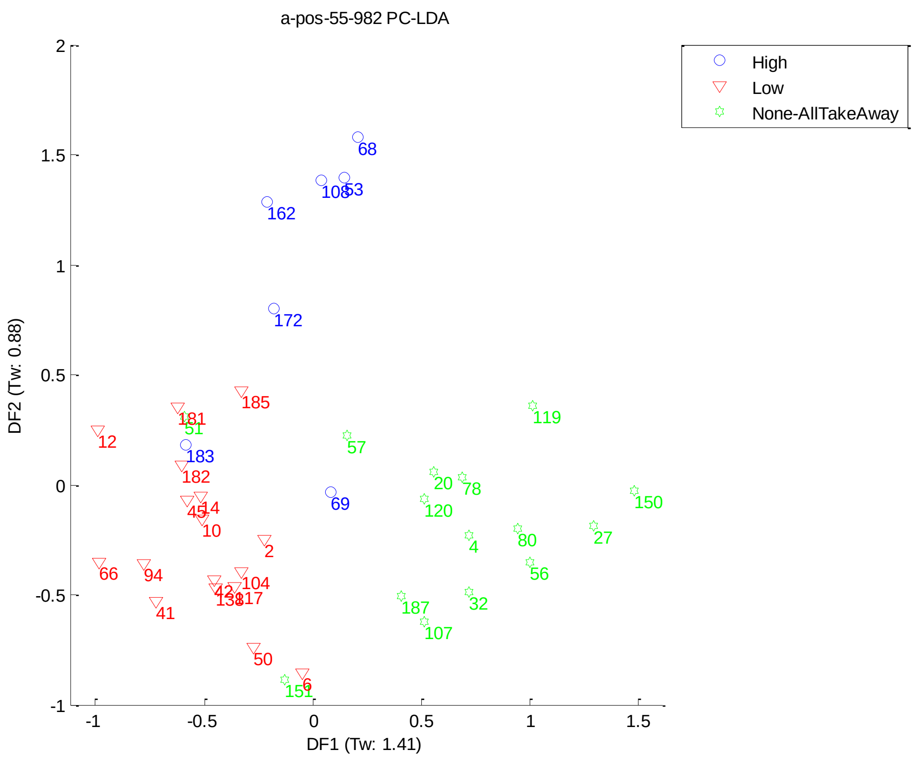
PCA plot Negative mode m/z 57-873 for pizza



PC-LDA plot Negative mode m/z 57-873 for pizza

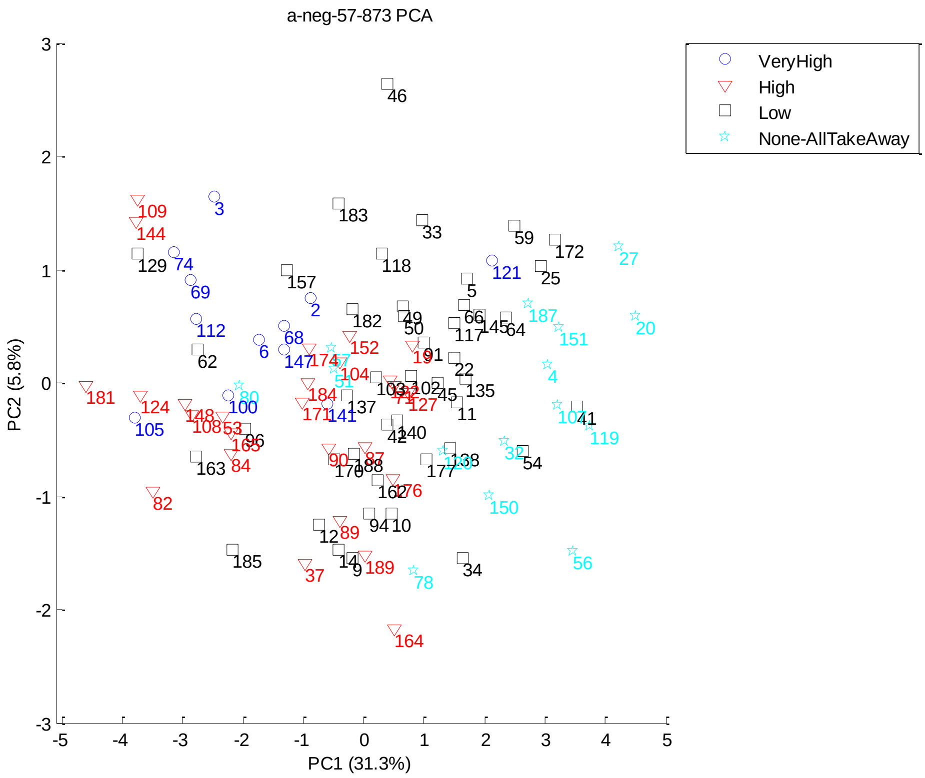


PCA plot Positive mode m/z 55 - 982 for pizza

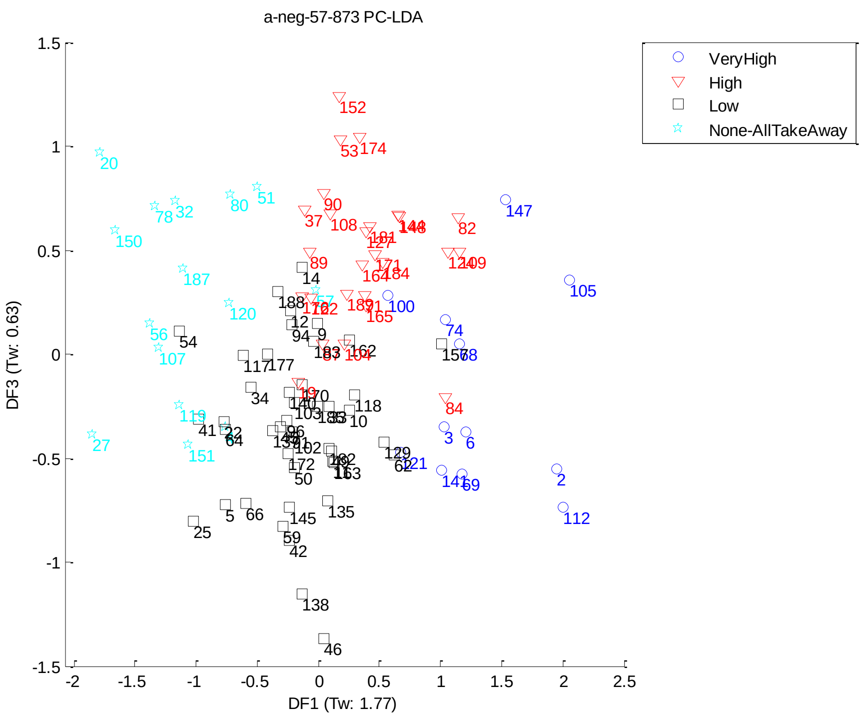


PC-LDA plot Positive mode m/z 55 - 982 for pizza

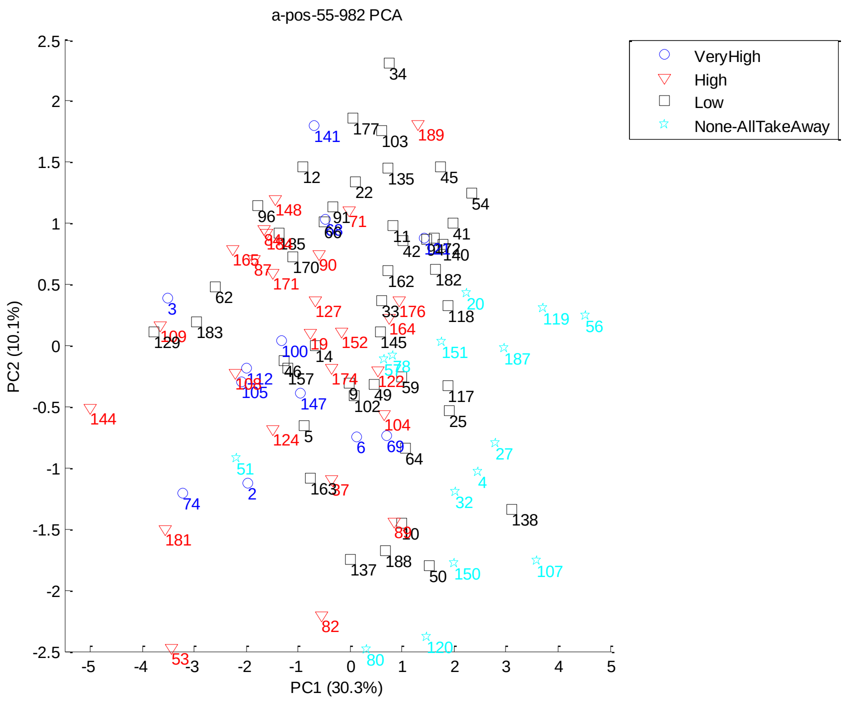
9.47 STUDY 3 METABOLITE FINGERPRINTS OF ALL TAKEAWAY FOOD



PCA plot Negative mode m/z 57-873 for all takeaway food



PC-LDA plot DF1 vs DF3 Negative mode m/z 57-873 for all takeaway food



PCA plot Positive mode m/z 55 – 982 for all takeaway food

9.48 PUBLICATIONS IN PRINT