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Glasgow Theses Service http://theses.gla.ac.uk/ theses@gla.ac.uk Dietary Glycaemic Index, Glycaemic Load and Insulin Resistance (HOMA_{IR}) of Healthy South Asians in Glasgow, UK

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A Thesis submitted to the University of Glasgow In fulfilment of the requirements for the Degree of Doctor of Philosophy

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ABSTRACT

High habitual dietary glycaemic index (GI) and glycaemic load (GL) may relate to elevated insulin resistance and therefore may be more important and relevant in South Asian populations known for high prevalence of insulin resistance. The main objective of this research was to investigate the dietary GI, GL and insulin resistance of a sample of healthy South Asians in Glasgow, UK (a total of 111 healthy individuals: 60 males, 30 South Asians and 30 Europeans; 51 females, 22 South Asians and 29 Europeans). Estimation of dietary GI and GL (from weighed food intake records) considered the GI values of single foods and mixed-meals from relevant publications and from laboratory food/mixed-meal GI measurements (Chapter 3). The GI of key staple South Asian foods alone (chapatti, rice, pilau rice) and as mixed meals with curried chicken was measured using standard methods on 13 healthy subjects. The key staples had medium GI (chapatti, 68; rice, 66 and pilau rice, 60) and glycaemic responses to the mixed-meal of staples with curried chicken were found to be lower than the staples eaten alone. GI of the mixed-meals fell in the low GI category (chapatti with curried chicken, 45 and pilau rice with curried chicken, 41). Weighed food intake records (WFR) (recorded for 3-7 days) and self-administered previously validated food frequency questionnaires (FFQ) (applied to habitual food intakes in the past 6 months) was assessed for agreement through correlation analyses, cross-classification analysis, weighted Kappa statistics and Bland and Altman statistics. The two methods mostly agreed in carbohydrate (CHO) food intakes implying that the WFR reflected habitual intakes (Chapter 4). In consideration of potential confounding effect of physical activity on the relationship between dietary variables and HOMA_{IR}, physical activity level (PAL) and Metabolic equivalent score (METS) of main daily activities of study subjects were derived from self-reported physical activity records (Chapter 5). Mean PAL were similar between South Asian and European males (median PAL of 1.61 and 1.60, respectively) but South Asian females tended to be less physically active than European females (mean PAL of 1.57 and 1.66, respectively). South Asians were less physically active in structured exercise and sports activities, particularly South Asian females and South Asians (males and females combined) with reported family history of diabetes showed inverse relationship between daily

energy expenditure and HOMA_{IR}. South Asians were found to be more insulin resistant than Europeans (HOMA_{IR} median (IQR) of 1.06 (0.58) and 0.91 (0.47), pvalue= 0.024 respectively in males; mean (SD) of 1.57 (0.80) and 1.16 (0.58), pvalue= 0.037, respectively in females) despite similarities in habitual diet including dietary GI and GL. The mean habitual dietary GI of South Asians was within the medium GI category and did not differ significantly from Europeans. South Asian and European males' dietary GI (mean, SD) was: 56.20, 2.78 and 54.77, 3.53 respectively; p-value=0.086. South Asian and European females also did not differ in their dietary GI (median, IQR) was: 54, 4.25 and 54, 5.00; pvalue=0.071). Top three staples ranked from highest to lowest intakes in the South Asian diet were: unleavened breads (chapatti, Naan/Pitta, Paratha), rice, bread (white, wholemeal, brown), and potatoes. After statistically controlling for energy intake, body mass index, age, physical activity level and sociodemographic status, an inverse relationship (Spearman partial correlation analyses) between dietary GI and HOMA_{IR} was observed (r, -0.435; *p*-value, 0.030) in South Asian males. This may be explained by the observation that the lower the dietary GI, the lower also, the total carbohydrates and fibre intakes and the higher the fat intake. In South Asian females, dietary GI and GL respectively, did not relate to $HOMA_{IR}$ but sugars intake related positively with HOMA_{IR} (r, 0.486; *p*-value, 0.048). South Asian females, compared to European females, reported higher intakes of dietary fat (38.5% and 34.2% energy from fat, respectively; p-value=0.035). Saturated fatty acid (SFA) intakes did not differ between ethnic groups but SFA intakes were above the recommended level of 10% of total dietary energy for the UK in all groups, the highest being in SA females. In conclusion, Ethnicity (South Asian), having family history of diabetes, the wider diet profile rather than habitual dietary glycaemic index and glycaemic load alone (low GI, low fibre and high fat diets in males for instance; and high fat, high sugar diets in females) as well as low physical activity particularly in structured exercise and sports may contribute to insulin resistance in South Asians. These observations should be confirmed in larger future studies.

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DECLARATION OF PUBLICATION

The following are publication and abstract arising from work associated with this thesis:

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Thank you and God bless and reward you all.

AUTHOR'S DECLARATION

I declare that the thesis is the result of my own work, Ramlah George @ Mohd Rosli, except where explicit reference is made to the contribution of others. I declare that this thesis does not include work forming part of a thesis presented successfully for another degree.

STATEMENT OF COLLABORATION

The author acknowledges contribution made by others in the conduct of the work for this research. Afia Aslam was involved in recruitment of a proportion of the South Asian subjects that had participated in this research.

LIST OF ABBREVIATIONS

BMI	body mass index
BMR	basal metabolic rate
CRP	C-reactive protein
Diabetes T2	diabetes type 2
CV	coefficient of variation
CRF	Cardiorespiratory fitness
СНО	carbohydrate
E	energy
EE	energy expenditure
E _{rep}	reported energy intake
EI	energy intake
FFQ	Food Frequency Questionnaire
GI	glycaemic index
GL	glycaemic load
HOMA _{IR}	Homeostasis Model Assessment of Insulin Resistance
IAUC	incremental area under the curve
MET	metabolic equivalent
METs	metabolic equivalent score
NMES	non-milk extrinsic sugars
PAL	physical activity level
RMR	resting metabolic rate
SD	standard deviation
SE	standard error of mean

Chapter 1 Introduction and Literature Review

1.1 Introduction

The aim of this chapter is to provide background to the studies presented in this thesis by providing relevant scientific evidence which will establish the rationale and framework for these studies. The review begins with the definition and concept of glycaemic index (GI) and glycaemic load (GL) as well as its influencing factors and the GI of South Asian foods/mixed-meals. This is followed by background on insulin resistance, with emphasis on risk and influencing factors such as genetic predisposition, physical activity and the diet as well as prevalence of insulin resistance in South Asians. Then evidence is provided from previous studies which investigated relationship between dietary GI and GL and health particularly on dietary GI, GL and insulin resistance. Lastly the South Asian diet will be reviewed which focuses on the overall diet and food/eating habit of South Asians as well their diet in context of health and disease.

1.2 Glycaemic index and glycaemic load

1.2.1 Definition and concept of glycaemic index and glycaemic load

Glycaemic index (GI) describes the blood glucose response after consumption of a carbohydrate containing food item (the test food) in comparison to a carbohydrate containing reference food (glucose or white bread) measured under standard conditions (FAO/WHO, 1998). The glycaemic index (GI) concept was first introduced by Jenkins et al. (1981) who determined the physiological response, specifically blood glucose response, of healthy individuals to some commonly eaten foods to supplement food composition tables based exclusively on chemical analysis. The GI of the food is defined as the incremental area under the blood glucose response curve (AUC) of a 50g available carbohydrate portion of a test food consumed by an individual under standard conditions expressed as a percentage of the AUC following consumption of a reference food (50 g of either a glucose solution or white bread) consumed by the same person on a different day (FAO/WHO, 1998). The GI classifies foods quantitatively according to the blood glucose-raising potential of the carbohydrates in the foods or the rate of carbohydrate absorption as shown in the glucose response (Jenkins et al., 1981). Foods are classified as either having low (<55), medium (55-69) or high (\geq 70) GI, where glucose = 100 (Brand-Miller et al., 2003). The principle of the GI is that the slower the rate of carbohydrate absorption, the lower the rise of blood glucose level and the lower the GI value.

The GI relates to a standard quantity of carbohydrates in a particular food but in practice, the usual servings consumed for different foods and meals vary. To account for both the quality (GI) and quantity of carbohydrates, a measurement known as the glycaemic load (GL) was introduced. GL allows comparisons of how average or realistic portions of different foods will likely affect blood glucose (Salmeron et al., 1997c). The GL assesses the total glycaemic effect of the diet and it is calculated by multiplying the amount of carbohydrates (in grams) provided by a food by the GI of the food, divided by 100 (Willett et al., 2002). A GL of 20 or more is considered high, 11 to 19 are medium, and 10 or less is low.

1.2.2 Glycaemic index and glycaemic load: influencing factors

The glycaemic response to foods rely on the rate of gastric emptying, the rate of digestion and absorption of carbohydrates from the small intestine (Jenkins et al., 1987) as well as on the effects of other food factors on non glucose mediated insulin secretion (Ostman et al., 2001). Several other factors that influence glycaemic index of foods have been investigated and these factors explain the wide variations in published glycaemic index values for similar food items.

It has been hypothesized that the metabolic effect of low-GI foods relates to the rate at which carbohydrates are absorbed from the gut. Low-GI foods are characterized by the slower rate of carbohydrate absorption resulting in a lower rise in blood glucose levels. To confirm this hypothesis, insulin secretion and serum free fatty acid were shown to be reduced when glucose was sipped at an even rate of 180 min compared with the same quantity of glucose consumed as a bolus at zero time (Jenkins et al., 1990).

Macronutrients and fibre

Fat and protein showed negative association with GI (Jenkins et al., 1981) and coingestion of protein and fat (0-30g) with carbohydrate (50 g of glucose dissolved in water) reduced glycaemic responses independently from each other in a linear, dose-dependent way (Moghaddam et al., 2006). The GI lowering effect of fat and protein may be attributed to their role in delaying gastric emptying thus delayed glucose absorption (Collier and O'Dea, 1983). A carbohydrate meal (50g potato) eaten together with relatively large amounts of fat (50g) and protein (50g of low fat veal) significantly increased gastric inhibitory polypeptide responses and lowered the postprandial glucose response of 8 normal study participants. Insulin response however, were not reduced (Collier and O'Dea, 1983). Lower amounts of fat (8-24g) added in mixed meals containing 38-104 g carbohydrate had little effect on predicted glycaemic response (Wolever and Bolognesi, 1996).

Earlier studies did not find correlations between GI and dietary fibre (Jenkins et al., 1981) because it was suggested that many of the high fibre foods that were studied comprised of wheat products known to have small effect on blood glucose. Later studies which investigated many more types of carbohydrate rich foods found an inverse relationship between total dietary fibre, namely insoluble fibre, and GI (Wolever, 1990). Soluble fibre such as guar gum, when added to a carbohydrate food has also been shown to reduce glycaemic response (Nuttall, 1993) as well as high levels of beta-glucan fibre (Jenkins et al., 2002). Viscous fibre such as guar and pectin appear to lower postprandial glucose and insulin levels in a certain degree due to a slower gastric emptying rate and slower movement towards the site of absorption (Torsdottir et al., 1991). Naturally occurring levels of viscous fibre in common cereals such as white bread and wholemeal bread only have small impact on glycaemia response but dietary fibre as part of an intact botanical structure, as in barley kernels and pumpernickel bread, may be effective in reducing glycaemia. Legumes for instance, are rich sources of viscous dietary fibre which have low GI (Jenkins et al., 1981;Bjorck et al., 2000).

Starch

Fractions of starch such as amylose and amylopectin have different glucose lowering effects. In barley flour based bread of varying amylose content, the GI was found to decrease as the percentage of amylose in bread increased (Akerberg et al., 1998). This is because amylose gives a lower GI compared to amylopectin (Bjorck et al., 2000) due to its unbranched or linear structure which is hydrolysed more slowly than amylopectin which has a branched structure.

White, high-amylose Bangladeshi rice variety BR16 has a low GI of 37 (Foster-Powell et al., 2002). Basmati rice has an intermediary GI in the range of 52-69 (Henry et al., 2005a) or a lower GI of 43 (Aston et al., 2008) depending on cooking time and strain. In contrast, Jasmine rice is high in amylopectin and therefore has a very high GI of 109 while broken rice has a GI of 86 as shown in the International Tables for Glycaemic Index of Foods (Atkinson et al., 2008).

Acidity

Addition of organic acids to cereal-based meals (white wheat bread with vinegar) lowered postprandial blood glucose and insulin. Inclusion of acids/salts lactic acid, acetic acid or the sodium salt of propionic acid lowered glycaemia and insulinemic response in healthy subjects (Ostman et al., 2006). The proposed mechanism for the propionic and acetic acids is a slower gastric emptying rate (Darwiche et al., 2001) and the lactic acid creates a barrier for the starch degrading enzymes (Ostman et al., 2001).

Mixed meal, Second-meal effect and meal frequency

The glycaemic index of a mixed meal can be calculated from the different proportions of each of the carbohydrate containing foods and their individual glycaemia index values. The glycaemia index for a mixed meal can also be directly measured from the area under the glucose response curve of the mixed meal. For example, when bread and beans are mixed in equal quantities, the resulting glycaemia response is midway between that of bread alone and beans alone (Wolever et al., 1985;Wolever and Jenkins, 1986). The glycaemia response to the same food or meal may be influenced by the time consumed and GI of a previous meal (second-meal effect) (Nilsson et al., 2008a;Nilsson et al., 2008b) and meal frequency (Jenkins et al., 1994).

Structure related factors and method of food processing and preparation Differences in physical and chemical characteristics of the foods influence GI. including the extent that food is chewed prior to swallowing (Read et al., 1986;Suzuki et al., 2005). Maintenance of high-starch crystallinity is an important factor in low-GI food. Preheated and flaked cereals have higher GI compared with less processed cereals. The GI increases as the degree of gelatinization increases in a product. Cellular structure or cell wall integrity is also important as GI increases with increased ripeness of fruits for instance. Formation of macromolecular interactions, and larger particle size distribution promotes lower GI (Bjorck et al., 2000).

Cooking method and food preparation

Cooking method and food preparation can modify the glycaemia index of foods. Highly processed convenience foods tend to have high GI while cooked pulse vegetables have low GI as their cell walls are resistant to cooking (Foster-Powell et al., 2002). The intact cereal grains of rye and granary bread all have low glycaemia indexes but when granary bread is processed to wholemeal bread, the grains are disrupted giving higher GI. Mashing of potatoes increase the GI by 25% (Pi-Sunyer, 2002) while pasta cooked al dente has lower GI than pasta that is cooked for a longer time possibly due to incomplete gelatinization and/or maintained physical structure.

1.3 Glycaemic index of South Asian foods/mixed meals

Main staples of South Asians such as chapatti/roti and rice have been published in the international table for GI (Foster-Powell et al., 2002;Atkinson et al., 2008) (Table 1.1) with varying GI depending on its type and source of carbohydrates. The GI of mixed meals rich in carbohydrates have been determined for several dishes namely chapatti served with bottle gourd and tomato curry (Chaturvedi et al., 1997) and lentil and curry with rice (Chew et al., 1988) as well as Indian flatbreads (rotis) prepared using whole wheat flour and 'atta mix' added whole wheat flour (Radhika et al., 2010). The GI of some traditional Indian vegetables have been studied. The vegetables were incorporated into mixed meals and served with chapati as traditionally eaten in Pakistan and the glycaemic response in nondiabetics and diabetics ranged from 29 to 103 and 25 to 94 respectively (Shoaib Akhtar et al., 2002).

Low-GI	Medium-GI	High-Gl
Chapatti, barley (42)	Chapatti, baisen (58)	Amaranth served with milk and nonnutritive sweetener (97)
	Chapatti, maize (62)	
Laddu		Chapatti, amaranth:wheat,50:50
(Fried dough, soaked in syrup) (27)	Chapatti, wheat,bottle gourd,tomato curry (66)	with bottle gourd and tomato curry (76)
	Chapati, wheat flour,thin, with green gram dhal (63)	Green gram dhal with varagu (78)
		Poori
	Dosai (parboiled and raw rice, soaked, ground, fermented and fried) with chutney (66)	(deep fried wheat flour dough)(70)
	Idli (parboiled and raw rice+black dhall, soaked, ground, fermented and fried) with chutney (69)	
	Pongal (rice and roasted green	
	gram dhal, press cooked) (68)	
Source: (Foster-	Powell et al., 2002)	

Table 1.1: Glycaemic index of	some South Asian foods
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1.4 Insulin resistance

1.4.1 Glucose and insulin

The maintenance or homeostasis of blood glucose levels is a key physiological function in mammals. Normal glucose homeostasis in humans is maintenance of blood glucose levels below 5.6mmol/L after an overnight fast (or after fasting for at least 10 hours). Prediabetic individuals have fasting blood glucose above 100 mg/dl, but not more than 126 mg/dl. Diabetes is diagnosed if fasting blood glucose exceeds 126 mg/dl (ADA, 2010). Because of the critical role of blood glucose homeostasis as a mechanism resulting in conditions such as diabetes, it is important to understand how glucose is regulated in humans and what specific problems at the cellular level could lead to impairments in the cellular and systemic mechanisms required for homeostasis of blood glucose.

Homeostasis of blood glucose is governed by the actions of insulin and glucagon (Giugliano et al., 2008) which involves coordinated functions of skeletal muscle, the liver, the endocrine pancreas, adipose tissue, and specific hypothalamic neurons (Henriksen, 2010). The liver contributes to regulation of glucose mainly through changes in hepatic glucose production. The adipose tissue acts as an endocrine organ that releases adipokines and is a site of insulin-dependent glucose disposal. The pancreatic α - and β -cells are the sites where insulin and glucagon are synthesized, respectively. The hypothalamus is also involved in the neural regulation of these organ systems.

The pancreatic α- and β-cells of the islets of Langerhans in the pancreas secrete the hormone insulin which plays a major role in controlling blood glucose. The insulin hormone acutely regulates glucose transport system in mammalian skeletal muscle through mechanisms involving several intracellular proteins that work in a specific sequential manner (Shepherd and Kahn, 1999;Zierath et al., 2000;Henriksen, 2002;Cartee and Wojtaszewski, 2007). Insulin directly influences cellular blood glucose concentration. After consumption of a meal containing carbohydrates for instance, glucose concentration in the plasma rises and this stimulates secretion of insulin. The insulin induces rapid entry of glucose into cells and cessation of glucose output by the liver, resulting in reduced concentration of glucose in the blood. This removes the stimulus to produce more insulin so insulin levels in the blood returns to its previous level. This demonstrates a negative feedback control of both glucose and insulin in the blood. An increase in plasma glucose concentration also inhibits the secretion of glucagons (a hormone secreted by the alfa cells of the islets of Langerhans in the pancreas). This regulates plasma glucose levels back to normal. In the fasting state, plasma glucose concentration decreases and this inhibits release of insulin while stimulates secretion of glucagons. Glucagon acts almost exclusively on the liver to increase hepatic glucose production.

Increased plasma concentration of free fatty acids and of certain amino acids also cause the release of insulin. The increase in insulin concentrations, in response to eating, which occurs before the rise in arterial glucose concentrations, is thought to be mediated largely via hormonal signals arising in the gastrointestinal tract (incretion effect). This means that insulin secretion will rise earlier (an anticipatory component) and to a greater extent than if plasma glucose was the only controller during ingestion of a meal. Another type of anticipatory regulation is how the parasympathetic neurons to the islets of Langerhans increase insulin secretion during ingestion of a meal. Exercise and stress, on the other hand, activate the sympathetic neurons and increase plasma epinephrine concentration, both of which inhibit insulin secretion.

1.4.2 Metabolic syndrome and insulin resistance

Metabolic syndrome is defined as a number of major metabolic disorders that predispose to cardiovascular disease (CVD) and type 2 diabetes mellitus. It is also known as the insulin resistance syndrome, syndrome X, dysmetabolic syndrome, or the deadly quartet (Grundy et al., 2004;Gallagher et al., 2010).

One factor that underlies metabolic syndrome is insulin resistance. Insulin resistance is a condition characterized by reduced action of the hormone insulin in activating the glucose transport system in skeletal muscle (ADA, 2010) it has also been defined as a state of impaired insulin action in insulin-dependent

tissues such as the liver, muscle and fat tissues (Withers et al., 1998). Insulin resistance results in reduction in insulin-stimulated glucose uptake in muscle and adipose tissue as well as impaired insulin-mediated suppression of hepatic glucose production and adipocyte lipolysis. When inadequate compensatory high levels of insulin (hyperinsulinemia) cannot overcome insulin resistance, this condition is called diabetes type 2 (ADA, 2010). Therefore insulin resistance is a known risk factor for the development of type 2 diabetes mellitus and cardiovascular disease (Lau et al., 2005).

Insulin resistance at the cellular level, may result from a disruption in any step of the insulin-signaling factors such as insulin receptor (IR), IR substrate (S)-1 or IRS-2, phosphoinositol-3-kinase (PI3K), protein kinase B (or phosphokinase B [PKB]/AKT), phosphoinositide-dependent kinase (PDK) and glucose transport (Rhee and Umpierrez, 2008). Predictors of insulin resistance like genetic predisposition or family history of diabetes type 2, obesity and lack of physical activity have been shown to be associated to defects in insulin signaling (see subtopic 1.4.4).

1.4.3 Risk factors and confounding factors for insulin resistance

Obesity

It has been suggested that obesity or adiposity is an important risk factor for diabetes, mainly because it can aggravate insulin-resistance (McKeigue et al., 1993;Banerji et al., 1999;Forouhi et al., 1999;Pomerleau et al., 1999;Chambers et al., 2001;Mohan et al., 2005). There is a positive relationship between body mass index (BMI) and adiposity. Migrant South Asians evidently have higher BMI compared to those living in their home country (Misra and Vikram, 2002;Misra and Vikram, 2004;Patel et al., 2006) and South Asians tend to possess greater central adiposity than Europeans (Mohan et al., 2005;Misra and Ganda, 2007). The cut-off points for obesity in different ethnic groups differ according to health risks seen at certain levels of BMI (Ntuk et al., 2014).

Insulin-resistant individuals who are obese or have a family history of type 2 DM have been shown to have decreased activation of an insulin signal called IRS-111

(Yu et al., 2002) which consequently caused reductions in insulin stimulated glucose transport (Rothman et al., 1995;Perseghin et al., 1996). It is suggested that all these can be caused by increased levels of free fatty acids (Shulman, 2000;Boden, 2003) and intracellular lipid content (Krssak et al., 1999;Perseghin et al., 1999) in insulin-resistant individuals.

British South Asian (158 (48.5% males)) volunteers aged 25-75 years with newly diagnosed impaired glucose tolerance or Type 2 Diabetes Mellitus were observed to have adverse adipocytokine profile which worsens with impaired glucose regulation. Insulin resistance is inversely associated with adiponectin independent of body mass index (BMI) and waist circumference in south Asians, implying that adipocytokine interaction contributes to the pathogenesis of metabolic disease in South Asians (Webb et al., 2012). Weight loss is associated with an increase in insulin sensitivity (McLaughlin et al., 2001) possibly by activating AMPK and restoring adipokine levels toward those associated with better insulin sensitivity. Adiposity is a major contributor to the difference in insulin resistance between rural and urban Indian men (age 30-50 years), in India; there was no additional contribution from inflammation or central obesity (Yajnik et al., 2008).

Diet

Diet is a modifiable factor that can influence health and disease. There is a wide body of evidence regarding dietary fat as it has been shown to contribute to the development of insulin resistance (Lovejoy et al., 2001;Wang et al., 2003;Ralston et al., 2013). The type of fat has also been shown to influence insulin resistance differently. High circulating levels of saturated fatty acids (SFA) and n-6 polyunsaturated fatty acids (PUFA) for instance, are associated with elevated fasting levels of insulin and glucose (Ebbesson et al., 2010;Rasic-Milutinovic et al., 2012;Zulyniak et al., 2012).

A high plasma free fatty acids (FFA) concentration is associated with an increased risk of developing diabetes (Paolisso et al., 1995) possibly by reducing insulin secretion (Zhou and Grill, 1994;Carpentier et al., 1999) and insulin action (Boden et al., 1994) which are factors involved in development of diabetes Type

2 (Ferrannini, 1998). Furthermore, elevated plasma TAG (the principal lipid abnormality in MetS) is an independent risk factor for CVD (Steinberg et al., 1996;Austin et al., 1998). TAG-rich lipoproteins may potentially have adverse effects on artery wall. High level of plasma TAG also increases atherogenicity of other lipoproteins. This reduces HDL (which is cardioprotective) and increases LDL, all of which are associated with CVD risk and MetS (Griffin, 1999).

Certain types of n-3 PUFA such as α-linolenic acid (ALA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) have been shown to improve glucose homeostasis and provide protective effects against the development of IR (Rasic-Milutinovic et al., 2012). The mechanism for this is by increasing production of insulin-sensitizing adipokines (e.g. adiponectin) and reducing the proinflammatory state of adipocytes and macrophages (Muramatsu et al., 2010;Oliver et al., 2012;Tishinsky et al., 2012). Another mechanism by which n-3 PUFAs improve insulin sensitivity is that enhanced intake of n-3 PUFAs change the quality of cellular membranes which increase insulin sensitivity (Flachs et al., 2014).

Consumption of energy dense/high fat diets is strongly and positively associated with overweight and insulin resistance, particularly when the excess body fat is located in abdominal region (Bray, 2004;Riccardi et al., 2004;Jimenez-Gomez et al., 2014). The link between fat intake and overweight is not limited to the high-energy content of fatty foods, but has also been shown to be associated with a reduced capacity to oxidize dietary fat (Astrup et al., 1994;Weinsier et al., 1995;Carstens et al., 2013;Jimenez-Gomez et al., 2014). Low-fat, high complex carbohydrate diet, compared to high saturated fat and baseline Spanish habitual diet, was shown to reduce insulin resistance and improve insulin signaling in subcutaneous white adipose tissue of MetS patients (Jimenez-Gomez et al., 2014). A high fiber diet may be protective against chronic diseases (Eshak et al., 2010;Kokubo et al., 2011;Wallstrom et al., 2012). The type of fiber consumed, whether soluble or insoluble could have different benefits. Higher dietary fiber is inversely and independently associated to incidence and risk of stroke in general population (Casiglia et al., 2013).

Physical activity and cardiorespiratory fitness

Physical activity is defined as "any bodily movement produced by skeletal muscles that results in energy expenditure" while physical fitness is "a set of attributes that people have or achieve" and the state of being physically fit has been defined as "the ability to carry out daily tasks with vigor and alertness, without undue fatigue and with ample energy to enjoy leisure-time pursuits and to meet unforeseen emergencies" (Caspersen et al., 1985).

Cardiorespiratory fitness (CRF) is a component of physical fitness and defined as "the ability of the circulatory, respiratory, and muscular systems to supply oxygen during sustained physical activity" (Lee et al., 2010) and includes maximal oxygen uptake (VO_{2max}) as measured by exercise tests such as treadmill or cycle ergometer. Sedentary behavior is defined as engaging in activities at the resting level of energy expenditure which includes sleeping, sitting, lying down, computer time, and viewing television (Janssen and Ross, 2012).

Physical activity and fitness strongly influences glucose tolerance and insulin resistance (Gill, 2007;Leite et al., 2009). Physical activity is also a strong predictor of diabetes risk and all-cause CVD mortality risks in both men and women(Lee et al., 2010). Fitness (VO_{2max}), for instance, has been shown to predict serum levels of insulin and glucose uptake and strongly positively correlate with insulin sensitivity and secretion in lean and healthy individuals (Larsen et al., 2012b). Furthermore, participating in 3 h/wk of vigorous-intensity activity was shown to be associated with a 22% lower risk of Myocardial Infarction among men which was partially explained by the beneficial effects of physical activity on HDL-C, vitamin D, apolipoprotein B, and hemoglobin A1c (Chomistek et al., 2011).

Notably, aerobic exercise and resistance training have been shown to improve insulin sensitivity in diabetic individuals (Goodyear and Kahn, 1998;Toledo et al., 2007) by increasing glucose transport into muscle (Goodyear and Kahn, 1998) and (Sakamoto and Goodyear, 2002). Because of these benefits of physical activity on blood glucose regulation, physical activity along with weight loss, are primary strategies of diabetes management (ADA, 2008).

Further evidence of the impact of physical activity on health and disease in that total sedentary time was found to be associated with increased cardiometabolic risk, independent of leisure time physical activity and adiposity in the The U.S. National Health and Nutrition Examination Survey (NHANES) study and in the Australian Diabetes, Obesity and Lifestyle study (Bankoski et al., 2011).

Furthermore, aerobic exercise has been suggested as an effective mechanism for reducing body weight and improving CVD (Thompson et al., 2003) and metabolic syndrome (Janssen and Ross, 2012). It has been suggested or rather assumed that regardless of the type of aerobics exercise, the exercise would affect these health parameters similarly given the same intensity, duration and frequency of exercise (Cox et al., 2010) but it is also possible that different modes of aerobic exercise may influence health variables differently.

1.4.4 Measurements for insulin resistance- Homeostatic Model of Assessment of Insulin Resistance (HOMA_{IR})

The gold standard technique for assessment of insulin sensitivity is the hyperinsulinemic euglycemic clamp (DeFronzo et al., 1979;Wallace et al., 2004;Matsuda, 2010) which is a method that uses direct insulin infusion as opposed to indirect methods that measure insulin concentration instead of infuse insulin such as homeostatic model assessment (HOMA_{IR}), fasting glucose/insulin ratio (FGIR), and quantitative insulin sensitivity check index (QUICKI) (Matsuda, 2010).

Several methods such as fasting or "homeostatic" models have been proposed due to relative ease of administration and less invasive technique. These include homeostatic model assessment (HOMA_{IR}), fasting glucose/insulin ratio (FGIR), and quantitative insulin sensitivity check index (QUICKI). These methods has been shown to correlate well with the standard hyperinsulinemic euglycemic clamp (Matthews et al., 1985;Laakso, 1993;Quon, 2001;Lorenzo et al., 2010).

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The insulin resistance index (HOMA_{IR}), has been widely used for the approximation of insulin resistance or as a surrogate measure of insulin resistance. HOMA_{IR} has particularly been employed in epidemiological research and clinical practice due to its ease of measurement of fasting plasma glucose and fasting plasma insulin concentrations. The original HOMA_{IR} model was developed in 1985 with a formula for estimation of insulin sensitivity from fasting serum insulin and fasting plasma glucose (Matthews et al., 1985). This formula (HOMA_{IR} = (fasting plasma insulin-fasting plasma glucose)/22.5) is simple and has been widely used in epidemiologic studies. Low HOMA_{IR} values indicated high insulin sensitivity, whereas high HOMA_{IR} values indicated low insulin sensitivity. The 75th percentile of HOMA_{IR} is frequently used as a cut-off for insulin resistance.

The physiological basis for the HOMA_{IR} model was previously described by Wallace et al. (2004) where it was mentioned that HOMA_{IR} is used to estimate insulin sensitivity and β -cell function from fasting plasma insulin and glucose concentrations. The relationship between glucose and insulin in the basal state reflects the balance between hepatic glucose output and insulin secretion, which is maintained by a feedback loop between the liver and β -cells. Predictions used in the model were from experimental data in humans and animals. In population studies, insulin resistance is often defined as greater than or equal to the 75th centile of the surrogate IR-measure distribution in subjects without diabetes mellitus (Balkau and Charles, 1999;Rutter et al., 2008). HOMA_{IR} values between 1.21 and 1.45 have been reported for normal subjects by Matthews et al. (1985). Large epidemiologic studies have used HOMA_{IR} to assess insulin sensitivity and report HOMA_{IR} to be approximately 2.6 on the basis of the 75th percentile (Hanson et al., 2000;Yeni-Komshian et al., 2000;Ascaso et al., 2003).

There are some limitations of surrogate measures of insulin resistance such as HOMA_{IR} (Buchanan et al., 2010; Thompson et al., 2014). HOMA_{IR} does not reflect the true function of dynamic ß-cell insulin secretion. HOMA_{IR} is only a prediction/approximation of glucose-stimulated insulin secretion from fasting steady-state data (Muniyappa et al., 2008) and the precision of HOMA_{IR} depends on the number of times fasting blood samples are obtained as well as the types

of insulin assays used in analysis (Bonora et al., 2000;Wallace et al., 2004). Although, single sample measurement of HOMA_{IR} has been applied in many population based studies and deemed acceptable for the purpose of obtaining population estimates (Wallace et al., 2004). In theory, however, insulin secretion is pulsatile therefore the mean of three samples taken at 5-min intervals to calculate HOMA_{IR} is more accurate than a single sample (Matthews et al., 1985).

HOMA_{IR} may not be appropriate to be applied in individuals with certain health conditions such as those with severely impaired or absent ß-cell function (Muniyappa et al., 2008) and in women with polycystic ovary syndrome (PCOS) (Diamanti-Kandarakis et al., 2004). In addition, HOMA_{IR} may not reflect beta-cell health in patients treated with sulfonylurea drugs (Wallace et al., 2004;Pfutzner et al., 2010). It may also not be appropriate to use surrogate measures of insulin resistance to quantify changes in insulin resistance over time (as in longitudinal data) because surrogate measures have been found to correlate poorly to direct measures of insulin resistance compared to when they are applied in cross-sectional settings. Surrogate measures possibly measure other factors besides insulin resistance (Hucking et al., 2008;Buchanan et al., 2010;Xiang et al., 2014).

Furthermore, the ability of surrogate measures of insulin resistance to predict insulin sensitivity has been shown to depend on race and gender. HOMA_{IR} for instance, may not be as reliable as other surrogate measures of insulin resistance such as Matsuda index and insulin sensitivity using oral glucose tolerance test in predicting insulin sensitivity in African Americans (Pisprasert et al., 2013). Therefore the application of HOMA_{IR} should also consider the race of the population studied.
1.5 Insulin resistance in South Asians (prevalence) and ethnic differences

Currently, diabetes Type 2 (T2D) is estimated to affect approximately 246 million people worldwide, with South Asians, particularly Asian Indians, having the highest and fastest growing prevalence. It has been established that South Asian ethnicity has been identified as a major risk factor for the development of diabetes T2. Central adiposity may also be a risk factor as well as insulin resistance and unhealthy lipid profile (Misra et al., 2010b;Garduno-Diaz and Khokhar, 2012). The risk of developing type 2 diabetes mellitus is especially high among both native and migrant South Asians (Bakker et al., 2013).

Furthermore, South Asians having the metabolic syndrome showed higher diastolic blood pressure, plasma triglycerides, fasting insulin and lower high-density lipoprotein-cholesterol (HDL-C) levels compared with UK Whites (Ajjan et al., 2007). The prevalence of the metabolic syndrome, CVD and type 2 diabetes is known to be higher in populations from the Indian subcontinent compared with the general UK population (Tillin et al., 2005).

Insulin resistance is highly prevalent in Asian Indian or South Asian migrants in Canada (Anand et al., 2000), United States (Enas et al., 1996; Mohanty et al., 2005), the United Kingdom (Samanta et al., 1991; McKeigue et al., 1992; McKeigue et al., 1993; Ramaiya et al., 1995) and in Europe (Chandie Shaw et al., 2002).

Many studies have reported that insulin resistance is more prevalent in South Asians than Europeans (McKeigue et al., 1988;McKeigue et al., 1991;Dickinson et al., 2002;Liew et al., 2003;Ehtisham et al., 2005;Barnett et al., 2006;Forouhi et al., 2006;Petersen, 2006;Nair et al., 2008;Ghouri et al., 2013;Goff et al., 2013). Ethnic groups living in the UK who originate from the Indian subcontinent, when compared with Europeans/white Caucasians, have higher levels of plasma triglycerides (TAG), insulin resistance, C-reactive protein, plasminogen activator inhibitor-1 and lipoprotein (a) and lower levels of HDL-cholesterol (Lovegrove, 2007;Merchant et al., 2007) higher glucose levels than Europeans(Forouhi et al., 2006;Gray et al., 2011;Ghouri et al., 2013) and substantially higher prevalence of diabetes in this ethnic group compared to Europeans (McKeigue et al., 1991). There is also a high prevalence of insulin resistance despite low rates of obesity among South Asians (McKeigue et al., 1992;McKeigue, 1996;Whincup et al., 2002). Diabetes Type 2 can appear nearly a decade earlier (McKeigue et al., 1988;Chowdhury et al., 2006). Diabetes Type 2 has also been shown to occur at a lower body mass index and waist circumference in South Asians than in Whites (Chowdhury et al., 2006;Mukhopadhyay et al., 2006). It has been suggested that recommended BMI threshold for obesity in South Asian populations should be lowered from 30 kg/m² to BMI 25 kg/m² (Misra et al., 2009a;Kumar et al., 2011;Tillin et al., 2015) and BMI 22 kg/m² (Ntuk et al., 2014) because of observations that South Asians, at a lower BMI, confer cardiometabolic-riskfactor profiles that are similar to those observed in populations of white European origin.

Observations of higher insulin resistance among South Asians than Europeans in Glasgow, UK have been reported (Ghouri et al., 2013). In the study by Ghouri et al., (2013), HOMA_{IR} and fasting glucose was found to be 67% (p<0.001) and 3% (p<0.018) higher, respectively, in South Asians than in Europeans. Ethnic differences in HOMA_{IR} were explained by total adiposity in South Asians and lower fitness levels (VO₂max). The authors concluded that lower cardiorespiratory fitness is the major factor associated with the higher insulin resistance and fasting glycaemia in middle-aged South Asian compared to European males in the UK.

1.6 Dietary glycaemic index, glycaemic load and insulin resistance

Diets with a high glycaemic index (GI) or glycaemic load (GL) have been hypothesised to increase the risk of diabetes, CVD and some cancers. This review will focus on GI, GL and its relation to diabetes and CVD risk. Two classic, landmark prospective epidemiologic cohort studies supported the hypothesis that habitual consumption of high glycaemic index diets are linked with an increased risk of developing diabetes type 2 in both women (Salmeron et al., 1997b;Schulze et al., 2004) and men(Salmeron et al., 1997a). These findings spurred on much research into this topic that researchers are still trying to confirm as there are many inconsistencies in study findings.

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Many studies that follow, both observational and intervention in design, have investigated dietary GI in relation to risk of chronic disease. Indeed the role of the glycaemic index (GI) and/or glycaemic load (GL) of foods in relation to health and disease has been a matter of debate (Hare-Bruun et al., 2008). Controlled dietary intervention trials on glycaemic index have supported the hypothesis that consumption of lower GI diet will favourably affect fasting blood glucose, glycated proteins, fasting insulin, insulin sensitivity and total cholesterol (Opperman et al., 2004; Livesey et al., 2008). There appeared to be evidence that the glycaemic index (GI) of dietary carbohydrates is important in disease prevention and control based on meta-analysis of studies that used randomised controlled trials and Chochrane review on studies that tested the effects of low GI diets on CHD (Brand-Miller et al., 2007;Kelly et al., 2008). Adding more weight to the beneficial effect of low GI diets on health was the meta-analysis of observational studies (Barclay et al., 2008) which concluded that low-GI and /or low-GL diets are independently associated with a reduced risk of certain chronic diseases such as diabetes and cardiovascular disease.

Later prospective cohort studies have shown associations between high GI and/or GL, with an increased risk for some (Ma et al., 2012; Sieri et al., 2013) but not all cardiovascular disease types (Fan et al., 2012). Similarly, low dietary GI and GL are associated with a reduced risk of type 2 diabetes in some (Jakobsen et al., 2010; Greenwood et al., 2013) but not all prospective cohorts (Simila et al., 2011). In contrast to this, evidence from randomised control trials has shown that low GI and GL diets are effective in reducing cardiovascular risk factors in overweight and obese subjects (McMillan-Price et al., 2006; Gogebakan et al., 2011; Schwingshackl and Hoffmann, 2013) and in subjects with type 2 diabetes (Jenkins et al., 2012). Two recent meta-analysis on observational studies have concluded that there appears to be good evidence of the association between dietary GI, GL and risk of type 2 diabetes (Dong et al., 2011; Greenwood et al., 2013). Inconsistencies in some study finding may be due to differences in study methods employed, underlying dietary patterns of food intake and limitations in dietary assessment methods and these limitations have been acknowledged by many investigators.

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1.6.1 Dietary glycaemic index and/or glycaemic load and insulin resistance $(HOMA_{IR})$

Insulin resistance is a known risk factor for the development of type 2 diabetes mellitus and cardiovascular disease (see subtopic 1.4.2), In light of this, an important question therefore arises as to whether the dietary glycaemic index or glycaemic load can affect certain risk factors of the metabolic syndrome, particularly insulin resistance. Several observational studies have addressed this question but findings are inconsistent. The following studies investigated the relationship between dietary GI, GL and HOMA_{IR} as a marker of insulin resistance.

Relevant observational, cross-sectional studies (that used HOMA_{IR} as a marker of insulin resistance) are summarised in Table 1.2. One large cross-sectional study in support of the beneficial effects of lower GI and GL diet with insulin resistance is by McKeown et al. (2004). This study involved a cohort of the Framingham Offspring Study totalling 2834 subjects (mean age of 54 ± 9.8 years). Usual dietary intake was assessed by self-administered, semiquantitative 126-item food frequency questionnaire (FFQ). After correcting for possible confounding effects of cereal fibre and whole-grain intakes, results showed that HOMA_{IR} (a measure of insulin resistance) increased significantly with an increase in both GI(p<0.001) and GL (p<0.03). Furthermore, the prevalence of the metabolic syndrome was found to be 41% higher in the group with the highest dietary GI intake (ranging 82-98 per day) compared to the group with the lowest GI intake (ranging <74 per day). GL was not significantly associated with prevalence of the metabolic syndrome.

A smaller cross-sectional study which examined data from two Dutch cohort studies (974 participants with mean age of 65± 9 years) (Du et al., 2008), found that GI significantly positively associated with HOMA_{IR} and fasting insulin after adjustment for potential confounders such as age, sex, smoking, physical activity, cohort, and the intake of total energy, alcohol, fiber, cholesterol, animal protein, plant protein, and saturated fatty acids. Interestingly, a 10-unit GI increment (corresponding to the difference between the lowest and the highest quintile), was significantly associated with a 23% increase in markers of insulin resistance. GI was also significantly inversely associated with HDL cholesterol. However, no association was evident between GL and any of the metabolic risk factors examined. In this study, habitual dietary intake was assessed by validated self-administered, quantitative food frequency questionnaire (FFQ) developed for the Dutch population.

Two other studies however, did not find association between daily dietary GI or GL with HOMA_{IR}. Lau et al. (2005), for instance, examined baseline crosssectional data from the Danish population-based Inter99 study which involved 5675 non-diabetic participants (aged 30-60 years). Habitual dietary intake was assessed using a 198-item food frequency questionnaire. No association was found between GI and HOMA_{IR} before and after adjustment for age, sex, smoking, physical activity, total energy intake, BMI, and waist circumference. They found that fibre intake and GL inversely associated with HOMA_{IR} before and after adjustment of the said confounders but GL no longer significantly inversely associated with HOMA_{IR} after fibre intake was accounted for. The authors suggested that GI or GL on its own does not influence HOMA_{IR} but that dietary fibre intake plays an important role in this.

Similarly,Liese et al. (2005), in the Insulin Resistance Atherosclerosis Study (IRAS) found no association between GI and insulin sensitivity. Although there was initially an inverse relationship between GL and insulin sensitivity, this was no longer apparent after adjustment for total energy intake. This study recruited 979 participants and used a 114-item interviewer-administered food frequency questionnaire for dietary assessment. These studies were carried out on a Western population and these findings may not be applicable to South Asians because of a difference in ethnicity, culture, food intakes, physiological profile and possibly in metabolisme of foods. Inconsistencies in study findings were probably due to underlying differences in the diet of the population studied, the dietary assessment method used (different types of FFQ) by which dietary GI and GL were derived from as well as variation in the estimation of GI from FFQs (how GI values were assigned to each item/group of foods in the FFQ list).

Author	Study design	Method (dietary assessment)	Parameters measured	Findings (after adjustment for confounding variables)
McKeown et al. (2004) n,2834 subjects; mean age: 54 ± 9.8 years	cross- sectional	FFQ; self- administered, semiquantitative 126- item	HOMA _{IR}	HOMA _{IR} increased significantly with an increase in both GI (p<0.001) and GL (p<0.03).
Du et al (2008) Dutch; n, 974 participants mean age: 65±9 years	cross- sectional	FFQ ; validated self- administered, quantitative,developed for the Dutch population	HOMA _{IR} ; HDL- cholesterol	-GI significantly positively associated with HOMA _{IR} and fasting insulin -GI was also significantly inversely associated with HDL cholesterol. -No association between GL and any of the metabolic risk factors examined
Lau et al. (2005) Danish ; n, 5675 nondiabetic participants aged 30-60 years	cross- sectional	FFQ, 198-item	HOMA _{IR}	-No association was found between GI and HOMA _{IR} -Fiber intake and GL inversely associated with HOMA _{IR}
Liese et al. (2005) n, 979 participants		FFQ , 114-item interviewer- administered	insulin sensitivity	-No association between GI and insulin sensitivity -No relationship between GL and insulin sensitivity

Table 1.2: Dietary glycaemic index, glycaemic load and insulin resistance studies (cross-sectional)

1.6.2 Dietary Glycaemic index, Glycaemic load and insulin resistance $(HOMA_{IR})$ in South Asians

There are limited publications on dietary GI and GL in South Asians. One study compared the dietary GI and GL of South Asian males and females, respectively with African males and females in Tobago (Wolever et al., 2002). The investigators hypothesised that Africans would have lower dietary GI than South Asians. African males were indeed found to have a lower dietary GI than South Asian males (56 ± 1 versus 59 ± 1, P < 0.05) but in females, no difference in their diet was observed. Correlations of diet variables to health parameters were not carried out in this study as it was aimed at providing diet information for the purpose of developing FFQ for future, larger studies in this population.

Dietary GI in the diet of South Asians in the UK could potentially be medium to high because of many sources of medium to fast-releasing CHO in their diet but it is could also be low due to concomitant amount of fat intake in their diet which could potentially be high. Adding fat to foods/meals has been observed to have lipid lowering effect (Collier et al., 1984;Moghaddam et al., 2006).

Currently, studies on GI and GL in relation to measures of insulin resistance such as HOMA_{IR} are very limited in South Asian population. This formed the basis for the study in Chapter 6 of this thesis where the dietary GI and GL of a healthy sample of South Asian males and females were investigated and related to fasting glucose, fasting insulin, insulin resistance indices (HOMA_{IR}).

One study on South Asians' dietary GL was by Radhika et al. (2009) who examined the relationship between dietary carbohydrates, glycemic load and high-density lipoprotein cholesterol (HDL-C) concentrations in Asian Indians. The study involved 2043 (886 men and 1157 women); individuals aged >20 years (mean age 40.2±12.3 years) who were randomly selected from Chennai Urban Rural Epidemiological Study (CURES), an ongoing population-based study on a representative population in India. The main findings were that total carbohydrates (P<0.001) and habitual dietary GL (derived from a validated semiquantitative FFQ) intake was inversely associated with plasma HDL concentrations where a stronger association was seen with dietary GL (P<0.0001) meaning that higher consumption of foods with high glycaemic load was associated with lower HDL-C and increased serum triglyceride. The pattern of decrease in HDL-C across the quintiles of GL (lowest to highest) was more obvious among men (1st vs 5th quintile: adjusted HDL-C: 4.3mg per 100 ml decrease (10.3%)) than women (1st vs 5th quintile: adjusted HDL-C: 3.2mg per 100 ml decrease (6.9%)). The authors suggested that this was evidence that both the quality and quantity of carbohydrates (GL) has significant effect on levels of HDL-C compared to the total amount of carbohydrates alone. In contrast, (Merchant et al., 2007) that also investigated South Asians but along with other ethnic groups, did not find an inverse association between HDL-C concentration and glycaemic load.

Another study on South Asians examined the association of dietary carbohydrates and glycaemic load, derived from validated meal-based semiquantitative FFQ), with the risk of type 2 diabetes among an urban adult Asian Indian population (Mohan et al., 2009) aged >20 years (n 1843; 771 men and 1072 women) who were randomly selected from CURES study, in southern India. Total carbohydrate (OR 4·98 (95% CI 2·69, 9·19), P,0·001), glycaemic load (OR 4·25 (95% CI 2·33, 7·77); P,0·001) and glycaemic index (OR 2·51 (95% CI 1·42, 4·43); P¼0·006) were associated with type 2 diabetes. Dietary fibre intake was inversely associated with diabetes (OR 0·31 (95% CI 0·15, 0·62); P,0·001).

These two studies are not comparable to other studies that have been carried out on Western populations. These studies were timely in that they investigated South Asians in India for which dietary GI and GL may be of more relevance since the diet of the South Asians in India were high in carbohydrates. These results may not apply to South Asians in the UK as those from India had carbohydrate intakes that were considerably high compared to the intakes of South Asians in the UK (approximately $65 \cdot 6\%$) (Mohan et al., 2009) versus 45% (Anderson and Lean, 2005) of total energy intake and their fat intakes also differed considerably (approximately 20 to 26% across quintiles of unadjusted dietary GI) (Radhika et al., 2009) versus 40% (Anderson and Lean, 2005).

1.7 South Asian Diet

In the United Kingdom, South Asians (individuals with ancestral origins from Pakistan, India, Bangladesh, and Sri Lanka) form the largest ethnic minority group comprising 6.8% of the total population in England and Wales (UK Census 2011) and 2.7% of the total population in Scotland (Scotland's 2011 Census).

Underlying genetic component (s) and environmental influences, for instance, sedentary lifestyle and carbohydrate rich diets are most usually cited as significant factors for higher incidence of Diabetes Type 2 in South Asians (Garduno-Diaz and Khokhar, 2012). The diet of South Asians has been studied in the UK and much of the focus of investigation in this population group has been on prevalence of chronic diseases and in understanding the tendency for them to have higher risks and prevalence of certain chronic diseases.

Nutrition studies on the South Asian diet have explored the eating habits of South Asians (Kassam-Khamis et al., 1996) and investigated possible links between food and nutrient intake and chronic diseases namely CHD (Anderson et al., 2005;Merchant et al., 2007) and diabetes (McKeigue et al., 1991). Some studies have compiled food composition tables for South Asians as well as developed and validated dietary assessment methods for this population(Kassam-Khamis et al., 1999;Judd et al., 2000;Kassam-Khamis et al., 2000;Sevak et al., 2004) and some have documented certain foods in the South Asian diet that South Asians perceive to be healthy (Pieroni et al., 2007) including medicinal perceptions of vegetables traditionally consumed by South-Asian migrants.

Health beliefs of UK South Asians and how these relate to lifestyle diseases have also been the subject of investigation (Ludwig et al., 2011;Lucas et al., 2013) focusing on diabetes (Grace et al., 2008;Lawton et al., 2008;Choudhury et al., 2009) and CHD (Farooqi et al., 2000;Darr et al., 2008).

The South Asian diet has been described as being rich in carbohydrates such as rice or/and unleavened bread (chapatti) as staples which are usually consumed with one or more type of curry (Mellin-Olsen and Wandel, 2005;Gilbert and

Khokhar, 2008; Wandel et al., 2008). Consumption of staples varied within subgroups of South Asians. Pakistanis for instance, ate more chapatti compared to Bangladeshis who ate mostly rice (Kassam-Khamis et al., 2000). This diet of curries and unleavened bread and/or rice as well as large amounts of fruits and vegetables indicates a dietary intake that is fairly high in fiber and moderate-to-low in fats (Wyke and Landman, 1997).

Asian Indians in India have been found to consume relatively more carbohydrates (60% to 67% of energy intake) (Shobana et al., 2007) as compared with the migrant Asian Indians in the United Kingdom (46% of energy intake) (Sevak et al., 1994) and the United States (56% to 58% of energy intake) (Kamath et al., 1999).

Dietary patterns may be associated with adverse metabolic changes which influence health. Pakistanis and Indians who live in Britain are 4-6 times more likely to develop diabetes type 2 than individuals of other ethnic groups in the general population (D'Costa et al., 2000) and this may be influenced by changes in lifestyle after migration (Bhatnagar et al., 1995). Lessons learned from the study on dietary habits of South Asians who migrated from their homeland to the UK was that upon migration, much of the traditional food habits were still maintained meaning that they're diets still included staples of carbohydrate such as breads (chapatti, roti, naan) and rice (Chadha et al., 1995) but some South Asians had also transitioned to less healthy aspect of Western diets like consumption of more processed foods especially in the younger generation (Gilbert and Khokhar, 2008).

It appeared that pre-migration diets that comprised of little meat and dairy products and large amounts of staples (chapattis, rice), pulses, fruit and vegetables had transitioned into diets that were higher in calories, fat, and sugar (Holmboe-Ottesen and Wandel, 2012) which were described as atherogenic (Anderson et al., 2005) as well as obesity and diabetes Type 2 promoting. Atherogenic diets ie. diets high in total fat and saturated fat, presented high coronary risk (Anderson et al., 2005). These dietary changes along with continued use of cooking methods such as frying and/or deep fat frying were suggested as reasons for why South Asians tended to have higher

percentage of energy from fat and saturated fat in comparison to Europeans (Anderson et al., 2005; Anderson and Lean, 2005). One such feature of the artherogenic diet was the consumption of dietary fat which was found to be high and remarkably similar figures (percent of fat contribution to energy) have been reported previously by different authors as summarised in Table 1.3.

Table 1.3: The dietary fat intake of South Asians

Author	% of food energy from fat (mean)
Anderson et al., 2005	Migrant South Asian: 42.4% British born South Asian:39.9%
Joshi and Lamb,2000	Vegetarians: 35% fat Nonvegetarians: 38% fat
Reddy and Sanders, 1992	Gujerati female Hindus:38% indigenous females: 40%
Sevak, McKeigue and Marmot, 1994 McKeigue and Chaturvedi, 1996	38% 38%

One study however, had observed improvement in several dietary practices among South Asians after immigration to Canada (Lesser et al., 2014) such as increase in fruits and vegetables intake and less consumption of high fat/fried food. In addition, immigrant South Asians reported healthier food preparation and cooking methods ie. less deep frying and more grilling/stir-frying. More than one third of respondents, however, also reported increased consumption of convenience foods, soft drinks and desserts or candy as well as an increase in eating out.

One interesting study suggested that dietary changes could be responsible for the increase in diabetes in children (aged 9 to 11) after moving from Pakistan to Bradford, UK. This notion was based on the observation that South Asian children in Bradford, UK (group1) consumed higher quantities of food that

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contained more fat, protein, carbohydrate, and sugar than then their counterparts in Pakistan (group 2) and from what their parents recalled from their diet when they were in Pakistan (group 3). The Bradford children tended to consume meat, fish, and fast food, but fewer vegetables and dairy products than the other groups. The childhood diet recalled by adults (group 3) was similar to that of children currently in Pakistan (group 2) (Edwards et al., 2006).

Imbalances in the diet of South Asians for particular macronutrients/nutrients have been associated to insulin resistance. These imbalances include high intake of total dietary fat, saturated fats, polyunsaturated fatty acids (PUFA), trans fatty acids and carbohydrates and low intake of monounsaturated fatty acid, omega-3 PUFAs and fiber (Sevak et al., 1994;Lovegrove, 2007;Misra et al., 2009b).

The findings of an epidemiological study (Patel et al., 2006) of CHD risk which compared nutritional intake, lifestyle, glucose tolerance and CHD risk of Indian migrants living in the UK and in India found that the Indian migrants possessed more metabolic risk factors compared to those living in India. Differences in the diet were also observed between the two groups and this led the authors of the study to hypothesise that dietary differences accounted for the higher frequency of metabolic CHD risk factors. It was proposed that greater metabolic- CHD risk amongst the migrant Indians was possibly due to dietary change which transitioned to higher intakes of energy and fat at the expense of carbohydrate. The authors of that study also proposed that their data suggest higher glucose excursions and unregulated non-esterified fatty acids (NEFA) metabolism in Indian populations (Patel et al., 2005). NEFA through its role on lipid metabolism and hepatic insulin resistance might be a driving factor in the higher CHD risk in South Asians (Hanley et al., 2002). However, it may also be possible that increased plasma NEFA is in response to insulin resistance and not the other way around (Olefsky and Glass, 2010).

In more current studies, dietary patterns have been shown to contribute to cardiovascular disease (CVD) risk in South Asians. One study in the United States on Asian Indians (n=150) aged 45 to 84 years, without known CVD observed that the Vegetarian diet, compared to the Western diet, was associated with lower

homeostasis model of assessment-insulin resistance (-1.12 mmol/L \times mU/L; P=0.05) and lower high-density lipoprotein cholesterol (-4.77 mg/dL; P=0.09). The authors concluded that healthy food/diet choices could help Asian Indians improve risk factors for CVD (Gadgil et al., 2014).

Carbohydrates, glycaemic index (GI) and glycaemic load (GL) of South Asian diet

Observations that support and reject the possibility that dietary GI and GL relate to health and disease was previously described (see subtopic 1.6). Studies on dietary glycaemic index and load of the South Asian diet in the UK are lacking but the GI concept may be of relevance to South Asians given that the traditional food habit of South Asians is built around CHO (carbohydrates) and that they are at higher risks of chronic diseases compared to other ethnic groups (Merchant et al., 2007). Although this has largely been attributed to genetic predisposition (Adair and Prentice, 2004) and body fatness (Gallagher et al., 2010;Webb et al., 2013); dietary factors may also be a contributing factor (see subtopic 1.4.3) including dietary GI and GL.

From the limited pool of publication for this specific topic, two big observational studies on South Asian population in India have investigated GI, GL in relation to CVD (see subtopic 1.6.2) and one small study by (Wolever et al., 2002) compared the dietary GI and GL of South Asian males and females, respectively with African males and females in Tobago. For the study by (Wolever et al., 2002), there was no significant difference in diet between AF and SA women. Intakes of energy, fat, protein, carbohydrate and fibre, and diet and glycaemic load also did not differ significantly. African males were found to have a lower dietary GI than South Asian males (56.0 ± 1.1 versus 59.0 ± 1.0 , P <0.05) but in females, no difference in their diet was observed (57.4 ± 1.1 verses 58.0 ± 0.9 , P > 0.05). Correlations of diet variables to health parameters were not carried out in this study as the aim was to obtain diet information to develop an FFQ for this population that will be used to test hypothesis for GI and diabetes in future. The investigators of the study concluded that these findings suggest that dietary GI may contribute to the development of diabetes Type 2.

The study by Radhika et al. (2009) examined the relationship between dietary carbohydrates, glycemic load and high-density lipoprotein cholesterol (HDL-C) concentrations in Asian Indians. The study involved 2043 (886 men and 1157 women); individuals aged >20 years (mean age 40.2 ± 12.3 years) randomly selected from Chennai Urban Rural Epidemiological Study (CURES), an ongoing population-based study on a representative population of Chennai city in southern India. The mean glycemic load was 386.8 ± 123.4 for men and 341.1 ± 111.4 for women (medium quintile). The mean dietary glycemic load intake ranged from 214 (lowest quintile) to >535 (highest quintile). Values for glycaemic index were not described by the authors.

The study by Mohan et al. (2009) examined the association of dietary carbohydrates and glycaemic load with the risk of type 2 diabetes among an urban adult Asian Indian population aged >20 years (n 1843; 771 men and 1072 women) who were randomly selected from the Chennai Urban Rural Epidemiology study, in southern India. For men, the median unadjusted total dietary carbohydrate intake was 406 (SD 117) g/d, GI was 69 (SD 3), GL was 277 (SD 86) and dietary fibre was 2.87 (SD 0.7) g/1000 kJ; for women, the corresponding values were 402 (SD 124) g/d, 69 (SD 2), 276 (SD 89) and 2.94 (SD 0.7) g/1000 kJ, respectively.

Suggested mechanism of dietary imbalances in the diet of South Asians that could lead to insulin resistance

As mentioned under subtopic 1.4.3, dietary fat has been shown to contribute to the development of insulin resistance (Lovejoy et al., 2001;Wang et al., 2003;Ralston et al., 2013). The type of fat has also been shown to influence insulin resistance differently. High circulating levels of saturated fatty acids (SFA) and n-6 polyunsaturated fatty acids (PUFA) for instance, are associated with elevated fasting levels of insulin and glucose (Ebbesson et al., 2010;Rasic-Milutinovic et al., 2012;Zulyniak et al., 2012).

The quality of dietary fats consumed, for instance, polyunsaturated fatty acids, monounsaturated or saturated fatty acids as well as amounts and ratios of different dietary fats influence fatty acid composition of cellular membranes (Misra et al., 2010a). Fatty acid composition of cellular membranes such as the ratio of polyunsaturated fatty acids to saturated fatty acids in membrane phospholipids in turn, relate to Insulin sensitivity (Weijers, 2012;Flachs et al., 2014). Furthermore, elevated plasma TAG (the principal lipid abnormality in MetS) is an independent risk factor for CVD (Steinberg et al., 1996;Austin et al., 1998). TAG-rich lipoproteins may potentially have adverse effects on artery wall. High level of plasma TAG also increases atherogenicity of other lipoproteins. This reduces HDL (which is cardioprotective) and increases LDL, all of which are associated with CVD risk and MetS (Griffin, 1999).

Certain types of n-3 PUFA such as α -linolenic acid (ALA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) have been shown to improve glucose homeostasis and provide protective effects against the development of IR (Rasic-Milutinovic et al., 2012). The mechanism for this is by increasing production of insulin-sensitizing adipokines (e.g. adiponectin) and reducing the proinflammatory state of adipocytes and macrophages (Muramatsu et al., 2010;Oliver et al., 2012;Tishinsky et al., 2012).

Consumption of energy dense/high fat diets is strongly and positively associated with overweight and insulin resistance, particularly when the excess body fat is located in abdominal region (Bray, 2004;Riccardi et al., 2004;Jimenez-Gomez et

al., 2014). The link between fat intake and overweight is not limited to the high-energy content of fatty foods, but has also been shown to be associated with a reduced capacity to oxidize dietary fat (Astrup et al., 1994;Weinsier et al., 1995;Carstens et al., 2013;Jimenez-Gomez et al., 2014).

Low-fat, high complex carbohydrate diet, compared to high saturated fat and baseline Spanish habitual diet, was shown to reduce insulin resistance and improve insulin signaling in subcutaneous white adipose tissue of MetS patients (Jimenez-Gomez et al., 2014). A high fiber diet may be protective against chronic diseases (Eshak et al., 2010;Kokubo et al., 2011;Wallstrom et al., 2012). The type of fiber consumed, whether soluble or insoluble could have different benefits. Higher dietary fiber is inversely and independently associated to incidence and risk of stroke in general population (Casiglia et al., 2013).

There are also studies that do not support the notion of "unhealthy diet or diet imbalances" in explaining metabolic disturbances in South Asians. This is mostly based on observations that South Asian diets have been found to be healthier in comparison to the diet of white Europeans (Sevak et al., 1994;Becker et al., 2006;Bowen et al., 2011;Leung and Stanner, 2011). South Asians compared to Europeans, for instance, had lower total dietary fat intake (36.5% vs 39.2% of energy intake) and higher dietary fibre intake (3.2 vs 2.0g/MJ) (Sevak et al., 1994).

Furthermore, South Asian communities in different regions and religion differ in their diet but despite this difference, South Asians, no matter where they are or what their diet, still had similar and noticeably higher prevalence of diabetes when compared to white Europeans. When habitual food/diet of individuals with different levels of glucose tolerance (normal glucose tolerance, impaired glucose tolerance, and newly diagnosed diabetes T2) was compared, no obvious difference in the diet was found (Simmons and Williams, 1997). This does not seem to support the suggestion that differences in diet explain for high prevalence of insulin resistance among South Asians. There is a possibility that it is not the diet per se but how individuals of different ethnicity respond to a particular diet. Nutrigenetics and nutrigenomics perhaps may play a role here.

1.8 Physical Activity of South Asians

Physical activity is a strong predictor of diabetes risk and all- cause and CVD mortality risks in both men and women (Lee et al., 2010) and the role of physical activity in reducing risk of non communicable diseases (Lee and Fujioka, 2011;Wen et al., 2011) particularly in South Asians (Ramachandran et al., 2006;Balagopal et al., 2008;Li et al., 2008) have been firmly established.

In the UK, South Asians have been shown to be less physically active than Europeans (Fischbacher et al., 2004;Yates et al., 2010;Williams et al., 2011a;Williams et al., 2011b;Ghouri et al., 2013). Physical activity levels of UK South Asians have been found to be low (Fischbacher et al., 2004;Williams et al., 2011a) particularly in South Asian women (Fischbacher et al., 2004). In addition, a consistently low level of physical activity in non-occupational activities such as sports was observed across the different subgroups of South Asians in the UK (Indian, Pakistani, Bangladeshi) compared to Europeans (Williams et al., 2011a). These findings were from 5421 South Asians and 8974 white participants aged 18-55 years who were participants of the Health Survey for England (1999-2004). South Asians were found to have considerably lower physical activity (by 60%) than Europeans even after controlled for age, sex, education, adiposity and self-reported health variations.

Apparent differences in physical activity of South Asians across subgroups (Indian, Pakistani, Bangladeshi) has also been observed (Rudat, 1994;Health Education Authority., 2000;Williams et al., 2011a) where Pakistani males reported the lowest levels of exercise followed by Bangladeshi men and then Indian men (Williams et al., 2011a).

In a Sri Lanka study, urban South Asian women (aged 30 to 45 years) were found to be at risk of dysglycaemia at lower levels of sedentary behaviour and greater physical activity than western populations (Waidyatilaka et al., 2013). In that study, physical activity and sedentary behaviours were assessed by the International Physical Activity Questionnaire (IPAQ).

1.9 Objectives of the chapters in this thesis

- 1. To determine the glycaemic index (GI) of chapatti, basmati rice, pilau rice, chapatti served with chicken curry and pilau rice served with chicken curry (Chapter 3) in order to provide GI values for calculation of habitual dietary GI and GL in the diet of South Asians (Chapter 6).
- To determine the agreement between weighed food records and food frequency questionnaires for carbohydrate food intakes by gender (males, females) and ethnicity (South Asian, European) (Chapter 4).
- 3. To determine and compare the physical activity level (PAL) of study subjects derived from self-reported physical activity records and METmin/day for five main activities (Chapter 5). The PAL of these subjects was considered as a potential confounding factor (controlled for in statistical analysis) in influencing relationship between diet and insulin resistance in South Asians (HOMA_{IR}) (Chapter 6).
- 4. To determine the habitual dietary glycaemic index and glycaemic load of a sample of South Asian males (n 30) and females (n 22) in Glasgow, UK and explain for this by describing their carbohydrate food intakes. Another aim was to determine whether dietary GI and GL related to HOMA_{IR} in South Asians through correlation tests (Chapter 6)

To make ethnic comparisons (South Asian and European males and females, respectively) of dietary glycaemic index (GI), glycaemic load (GL), other carbohydrate-related dietary variables and HOMA_{IR} (Chapter 6).

Chapter 2 General Methods

There are four study chapters in this thesis (Chapter 3, 4, 5 and 6). The first major study is presented in Chapter 3 while the second major study is presented in smaller study chapters (Chapter 4, 5 and 6). The methods shared by these studies are presented in this chapter while the methods that were specific to a particular study are described in the relevant chapter.

2.1 Ethics approval and study subjects recruitment

All studies were carried out with ethical approval of the Medical Faculty Ethics Committee, University of Glasgow. Study subjects were volunteers who were recruited from the local community in Glasgow, UK through advertisements posted at public places (Appendix I), social networks (Facebook), email and by word of mouth. Subjects were given the option to enquire about the study verbally (face to face or on the telephone) and through email. Individuals who came to the study site to enquire about the study were given a Subject Information Sheet for the particular study they enquired about (Appendix II) and given the opportunity to ask questions. Subjects were also verbally informed with full details of the study protocol that required their participation. All subjects that agreed to participate in the study gave their consent by signing a consent form for the study (Appendix III). They then completed a brief general questionnaire on demographic information and health screen questions (Appendix IV) to confirm that they were healthy.

2.2 Criteria of study subjects

The study subjects that were involved in the study in Chapter 3 were different from the study subjects that participated in the studies in Chapter 4, 5 and 6 (which shared the same subjects). There were specific inclusion and exclusion criteria for these two major studies. In general, all study subjects studies were healthy as defined by no diagnosed chronic diseases such as diabetes, heart problems, or other metabolic problems; not on any type of medication known to effect carbohydrate metabolisme, not pregnant, not breastfeeding and not vegetarian as confirmed through a General Questionnaire and Health Screen (Appendix IV). None of the study subjects were on any particular diet for medical reasons, weight gain or weight loss.

2.3 Anthropometric measurements

Subjects' height, weight, waist circumference and body fat percent were measured to provide information on their physical characteristics. Anthropometric measurements were conducted with subjects wearing light clothing and no shoes. Height was recorded to the nearest centimetre using a stadiometer (Holtain, UK), with subjects standing straight. Waist circumference was measured using a tape measure midway point between the iliac crest and the lower rib. Body weight (recorded to the nearest 0.1kg) and body fat percentage was measured using the Tanita BF-300 (Tanita UK Ltd, Yiewsley, Middlesex, UK). The scale was placed on a hard, level surface. Body mass index (BMI) was calculated using the standard formula: weight (kg)/height (m²).

Chapter 3 Glycaemic index measurement of key staple South Asian foods alone and as a mixed meal with Chicken Curry

The last two chapters provided a background on this overall research. In this Chapter, the glycaemic index (GI) measurement of some key staples alone and combined with curried chicken as a mixed meal is presented. This was carried out in consideration of the South Asian diet which included several staples in mixed meals for which the dietary GI was unknown. The GI of the foods measured in this study was subsequently used in the calculation of the dietary GI of South Asians and Europeans (Chapter 6). The study subjects were healthy South Asians residing in Glasgow, UK and this study was carried out in 2009-2012. The study participants for the GI measurement of South Asian foods study presented in this chapter were a different set of subjects from those who participated in the studies described in the chapters that follow in this thesis.

3.1 Introduction

The Glycaemic index (GI) of food is defined as the incremental area under the blood glucose response curve (AUC) of a 50g available carbohydrate portion of a test food consumed by an individual under standard conditions expressed as a percentage of the AUC following consumption of a reference food (50 g of either a glucose solution or white bread) over a period of two hours consumed by the same person on a different day (FAO/WHO, 1998). Jenkins et al. (1981) first introduced the GI concept from a study that investigated the physiological response; specifically blood glucose response, of healthy individuals to some commonly eaten foods to supplement food composition tables that only had chemical analysis information.

In general, estimation of average dietary GI from dietary records/surveys uses formulas that require investigators to know the GI value of foods reported by study subjects. In these types of studies, the GI values are usually obtained from published GI values for foods (Henry et al., 2005a;Henry et al., 2005b;Henry et al., 2006;Henry et al., 2007;Aston et al., 2008;Henry et al., 2008) and from the international tables for glycaemic index values (Atkinson et al., 2008). The reliability of these studies is largely dependent on the GI values used in calculation of average dietary GI. Although published GI values enable estimations to be made, these GI values are not exhaustive as they do not cover all foods that may be recorded in a dietary survey/record. Furthermore, published GI values for foods vary widely even for foods within the same category or that possess similar descriptions.

This wide range in the GI values of foods are caused by many factors such as type of carbohydrate, processing technique, addition of fat, protein and fibre to the foods etc (see details in Chapter 1). It has been recommended that foods, especially staples, be measured for GI in the place of origin where they were consumed where same brands, processing techniques and types of carbohydrates are more alike (Miller et al., 1992).

As previously mentioned, not all South Asian foods are included in GI tables and the impact of eating mixed meals may influence the GI of individual foods (Henry et al., 2006;Dodd et al., 2011). Therefore it was important to measure the GI of key staples and mixed meals in the diet of South Asians that were then used in determination of dietary GI and GL (Chapter 6). In this chapter, the GI value of two key staples in the South Asian diet namely chapatti and basmati rice were investigated as these staples contribute largely to carbohydrate intake in the South Asian diet. Although some GI values for chapatti and rice eaten alone or with curry have been published in the International Tables of Glycaemic Index and Glycaemic Load Values (Atkinson et al., 2008), the foods tested vary in source/brand, nutrient composition, ingredients, and preparation.

The GI of chapatti and basmati rice in this study was measured as they would normally be eaten in a mixed meal with curry. It was hypothesised that adding cooking oil, in the amount normally used by South Asians, to the staples in the form of a mixed meal such as chapatti or rice served with chicken curry, would significantly lower the GI because addition of oils/fats has been shown to decrease GI by delaying gastric emptying (Collier et al., 1984). Addition of toppings to foods (Henry et al., 2006) and varying amounts of fat/oil and protein to a meal have been found to be lower than the GI predicted (Dodd et al., 2011) based solely on the main carbohydrates foods/ingredients in the mixed meal. Direct measurement of the GI of the mixed meals would provide more reliable/accurate GI values for mixed meals. These GI values were then used in calculation of dietary GI and GL in South Asians and Europeans (Chapter 6). The GI values would also be useful in estimation of dietary GI in future studies particularly in populations that frequently consume these types of foods/meals.

3.2 Study objectives and Hypothesis

Objectives

- to estimate the GI of: chapatti, basmati rice, pilau rice, chapatti served with chicken curry (curry made to a typical South Asian recipe containing corn oil, chicken and tomatoes), and pilau rice served with curry and compare our results with published GI values
- 2. to assess the impact of mixed meals on GI; i) chapatti eaten alone versus chapatti eaten with chicken curry and ii) pilau rice eaten alone versus pilau rice eaten with chicken curry
- 3. to provide GI values for determination of dietary glycaemic Index and glycaemic load of South Asian and European subjects (Chapter 6).

Hypothesis

The study tested the hypothesis that:

- 1. chapatti and rice would be categorised as medium GI foods
- 2. the GI would decrease when these staples (chapatti and rice) are eaten with chicken curry as a mixed meal

3.3 Subjects

Ethical approval to carry out the study was granted by Medical Faculty Ethics Committee, University of Glasgow. Subject recruitment and inclusion criteria are described in Chapter 2, General Methods, Subtopic 2.1 and 2.2. A total of 15 healthy subjects (14 females and 1 male) participated in this study. The number of subjects recommended for studies in GI measurement of foods is at least 8 to 10 (FAO/WHO, 1998) or at least 10 (Brouns et al., 2005). Study subjects were aged 28 years (range 20-34) on average, had normal body mass index (BMI) with a mean of 21 kg/m² (range 18.5 -24.2 kg/m²), were non-smokers and had no intolerance/allergies to any of the test foods/mixed meals. Their average fasting blood glucose was 4.8mmol/L which is within the normal range for healthy people. Study subjects fulfilled the study subject criteria recommended (FAO/WHO, 1998) for the measurement of GI of foods.

3.4 Methods

3.4.1 Reference and test food

Glucose (Sigma-Aldrich, St. Louis, MO, USA) was chosen as the reference food because it is more easily standardized for international use (Wolever et al., 2003) than white bread which although more representative of a physiological meal (Wolever et al., 1991), has variation in both its composition and digestibility characteristics with when and where it is produced. Glucose was weighed (50 grams) and dissolved in 200ml of warm water.

Subjects ingested the reference food on two separate days and each test food was tested once, with a gap of at least one day between measurements to minimise carry-over effects. Although an average of three days for reference food testing would give a better average of the IAUC for glucose, the minimum of two days of reference food testing was employed as recommended by Brouns et al. (2005). This recommendation was derived from a simulation study conducted on the data of Wolever et al. (2003) which showed that the error of the estimate of mean GI was statistically significantly different only from one to two reference measurements and not for any other number of reference

measurement. The testing of the reference food twice instead of three times encouraged subjects to participate in this present study as it reduced the total number of visits to complete the study.

Five food items were tested for their GI based on glucose as the reference food. The test foods were key staples commonly consumed by the South Asian population group studied: chapatti, chapatti served with chicken curry, basmati rice, pilau rice, pilau rice served with chicken curry. Due to logistics and time constraints, only the most frequently consumed brand of chapatti and flour reported by South Asian subjects in food frequency questionnaires (FFQ) (Chapter 4) were selected for GI value measurement. All chapattis were made from chapatti flour (Elephant Atta Medium Chapatti flour, Rank Hovis Ltd., UK) and all rice dishes were of the basmati type (Laila Basmati Rice).

Portion sizes of test meals were calculated to provide 50 g available carbohydrate, according to manufacturers' nutrition information (FAO/WHO, 1998) and WinDiets 2005. In mixed meals, the ingredients were made up to provide 50 g of carbohydrate (Table 3.2). The composition differed depending on the ingredients used in the preparation of the foods. To provide 50g of carbohydrate, more whole wheat flour, in terms of dry raw weight (78g), was required to make chapatti compared to rice (65 g). Chapatti flour/atta and rice were almost similar in energy content. The addition of chicken curry to both chapatti and rice increased energy, macronutrient and fibre content. The test foods/meals recipes are described in Table 3.3. Preparation of all meals was standardised according to the manufacturer's instructions on the food packaging where applicable, compiled recipes from previous food intake studies on South Asians (Kassam-Khamis et al., 2000). All meals were prepared on the morning of the test and served warm.

3.4.2 Assignment of test foods to subjects

Healthy subjects (n=15) participated in this study. The testing of each test food involved 10 or 11 individuals but these individuals did not necessarily test all the test foods. For comparison of the staples eaten alone and in a mixed meal, the test meals were tested in pairs by 9 healthy subjects (pair 1-chapatti, chapatti served with curry; pair 2- pilau rice, pilau rice served with curry). This means each subject served as his/her own control and enabled the use of paired t-test analysis to determine if the mean GI value of the staple eaten alone differed from the GI value of the staple eaten in a mixed meal. A total of 6 subjects tested all 5 test foods, 3 subjects tested 4 test foods, 1 subject tested 3 test foods, 2 subjects tested 2 test foods while 3 subjects tested only 1 test food. Subjects were randomly allocated with test food (s) using Minitab (Minitab, Inc.). The reference food (glucose) was tested at the beginning and end of the test food measurements.

3.4.3 Glycaemic index measurement protocol

The protocol used to measure GI was adapted from that described by Wolever et al. (1991) and in accordance with procedures recommended by the FAO/WHO (1998). On the day before the test, subjects were asked not to consume any alcoholic beverages and caffeine containing drinks and to avoid intense physical activity for long periods of time (e.g. at the gym, excessive swimming, running, aerobics). Subjects were instructed to fast for at least 12 hours before the test but they were allowed to drink plain water in moderation. Subjects were studied in the morning after a 12 h overnight fast.

On the day of the trial, subjects were asked to relax (for at least 20 minutes) before their anthropometry measurements (height, weight, body composition) and waist circumference were taken. These anthropometry measurements were taken once during the subjects' first visit. Subjects were then encouraged to massage their hands and fingers to ensure good blood flow. Subjects' hands were kept warm with a fan heater. Subjects were asked to consume the reference/test food at a comfortable pace and finish within 15 minutes. Blood glucose measurements were taken before the subjects started eating the test or reference food (fasting) and over the next two hours (at 0, 15, 30, 45, 60,

90,120 mins). The test foods were served with 200 ml water. Another 200 ml of water was provided over the subsequent two hours. Subjects were encouraged to minimise physical activity during the testing.

3.4.4 Anthropometric measurements

Anthropometric measurements such as height, weight, waist circumference and body fat percent were taken in the morning on subjects wearing light clothing and no shoes using standard methods of measurement as described in Chapter 2, General Methods, Subtopic 2.4 Anthropometric measurements.

3.4.5 Blood glucose measurements

Standard methods were used to measure blood glucose (FAO/WHO, 1998). This standard GI measurement method allows choice of blood glucose sampling method (venous or capillary; whole blood or plasma) and glucose analysis method (eg. glucose hexokinase, glucose dehydrogenase or glucose oxidase method). Capillary blood sampling was carried out as recommended by (FAO/WHO, 1998; Wolever et al., 2003; Brouns et al., 2005) through finger prick as it has been suggested that capillary blood is more reliable than venous blood sampling (Wolever et al., 2003), improves sensitivity and removes the effect of potential changes in measured GI due to fluctuations in factors such as ambient temperature. Capillary blood have also been found to produce smaller variation (SD) in mean GI values compared to venous blood (Wolever et al., 2003) (FAO/WHO, 1998; Brouns et al., 2005). Blood glucose was measured using an automatic analyser, HemoCue Glucose 201+ Analyzer (HemoCue Limited) which was factory calibrated. The HemoCue glucose system has been found to correlate well with an established method of blood glucose analysis using Yellow Springs Instrument (YSI 2300 STAT; YSI) glucose oxidase analyzer (Stork et al., 2005).

Fasting capillary blood samples were taken before consumption of the food. Subjects were asked to consume the reference/test food immediately after this and additional blood samples were taken at 15, 30, 45, 60, 90 and 120 minutes after the test meal. Blood glucose readings were taken in duplicate per finger prick. A third reading was taken if the two initial blood glucose readings differed by more than 0.3mmol/L. Measurements were done under hygienic conditions. Before capillary blood sampling, subjects were encouraged to warm their hands to increase blood flow with the use of a fan heater. The following procedures for blood sampling by finger prick were recommended by Hemocue Limited. The middle or ring finger was usually used for sampling and the puncture site was slightly off centre from the central portion of the fingertip as these are generally less calloused, which facilitates puncturing and may be less painful. The finger was first cleaned with disinfectant and allowed to air dry completely to provide effective disinfection. The finger was lightly pressed from the top of the knuckle towards the tip then punctured with a lancet (Haemolance Plus safety lancets for normal flow, 21 gauge manufactured by Htl-Strefa S.A, Ozorków, Poland).

The first drop of blood was wiped away as it contains tissue fluid, which could cause specimen dilution, haemolysis and clotting. Excessive squeezing of the finger was avoided as this would cause tissue fluid to be expressed. The microcuvette (HemoCue Glucose 201+ Microvettes) was filled with blood in one continuous process and blood glucose was read within 40 seconds after filling. The principle of how these microcuvettes function is based on a glucose dehydrogenase method (HemoCue Limited, Angelholm, Sweden).

3.4.6 Quality control of analyser

The analyser was checked each time before testing using control solutions, Glucose Eurotrol GlucoTrol-NG (Eurotrol Inc., Burlington, MA, USA) as recommended by the manufacturer (HemoCue Limited, Angelholm, Sweden). The control solution (purified bovine plasma with glucose) level used in this study was 6.0mmol/l as this was relevant to the range of glucose concentration expected in this study. If the measured control solution differed from its known concentration, the HemoCue glucose analyser was checked according to the manufacturer's standard instructions and serviced accordingly.

3.4.7 Calculation of the glycaemic index

Data was analysed according to the method recommended by FAO/WHO (1998). For each subject and each test food/meal, the blood glucose measurements at each time point (0,15, 30, 45, 60, 90 and 120 minutes after the test food/meal) were plotted on a graph (blood glucose (mmol/L) vs time (mins)). The incremental area under the blood glucose response curves (IAUC) was then calculated geometrically by using the trapezoid rule. The area below the fasting baseline was not included in the calculation of IAUC. For each subject, the IAUC of the reference food (tested twice on separate days) were averaged and then the GI of the food were calculated by dividing the IAUC of the test food with the mean IAUC of the reference food multiplied by 100 (Equation 3.1). The GI of the test food was then calculated as the mean value across all subjects consuming that food.

3.4.8 Coefficient variation (CV) for glucose IAUC

The mean percentage of coefficient variation (CV%) for glucose IAUC was determined to assess the level of intra-individual (within the same subjects) variation of the reference (glucose) tests which were carried out twice by each subject. The CV% of glucose IAUC was calculated for each subject by dividing the standard deviation (SD) by the mean glucose IAUC multiplied by 100 and then averaged to obtain the mean CV% for the group of subjects.

Equation 3.1:

GI value of the test food = (mean IAUC value for the test food) X 100 (mean IAUC value for the reference food containing the same amount of CHO as in the test food)

GI, Glycaemic index IAUC, Incremental area under the curve CHO, carbohydrate

3.4.9 Handling of outlier values

Box-plot or box-and-whisker plots were used to identify any outlier values in the data. Any data not included between the whiskers were considered as an outlier. Mean GI values for subjects were widely distributed from the group mean, as shown by the standard deviation for the GI values, but no outlying data were observed.

3.4.10 Statistical analysis

Statistical analyses were performed with PASW Statistics 18 (SPSS Inc.). Data are represented as means with their standard errors unless stated differently. The normality of the data was tested using the Shapiro-Wilks statistics before carrying out further statistical analysis. All GI values for test foods and IAUC for glucose averages were normally distributed. One way analysis of variance (ANOVA) was used to compare the mean GI of chapatti, pilau rice and rice.

Paired t-test was used to determine if the mean GI was significantly different between the following foods: i) chapatti (alone) and chapatti served with curry and ii) pilau rice and pilau rice served with curry. Statistical significance was set at *P*-value of <0.05. The GI values of test foods were reported with their standard deviation (SD) and standard error of mean (SEM) values.

3.5 Results

3.5.1 Characteristics of the study subjects

The anthropometric and clinical characteristics of the study subjects are presented in Table 3.1. A total of 15 healthy subjects were studied, 14 females and 1 male, with mean age 28 years (range 20-34) and normal body mass index (BMI) mean 21 kg/m² (range 18.5 -24.2 kg/m²). The subjects' mean waist circumference was categorised as low risk. Their average fasting blood glucose was 4.8mmol/L which is within the normal range for healthy people. Study subjects fulfilled the study subject criteria recommended (FAO/WHO, 1998) for the measurement of GI of foods.

3.5.2 Coefficient of variation (CV) in glycaemic response to reference tests

The mean Incremental area under the curve (IAUC) for the reference food, glucose, was 278.1 mmol/min per litre (standard deviation (SD) 90.8; standard error of mean (SEM) 23.5). The mean intra-individual coefficient of variation (CV) in glycaemic response to the reference tests and SEM for the fifteen study subjects was $25.5\% \pm 1.7$ which are consistent with previously reported data (Wolever et al., 2003;Henry et al., 2005a).

3.5.3 Glycaemic response curves for test foods

Figure 3.4 show the glycaemic response curves for the five test foods in the healthy subjects. Each point represents the average blood glucose concentration for ten subjects at that time point measured at baseline (0 min) and subsequently at 15, 30, 45, 60, 90 and 120min after the start of eating the food. A comparison of the glycaemic response for all five test foods demonstrate the slower peak rise of blood glucose response, at 45 min, for chapatti and chapatti served with curry in comparison to pilau rice, pilau rice served with curry and plain rice which all peaked at 30 minutes.

The mean fasting blood glucose was similar before each test meal and the reference food (P=0.635) (Table 3.4). The mean IAUC for chapatti served with curry was significantly lower than for chapatti eaten alone (P=0.002) (Table 3.5)

and Figure 3.2). Similarly, mean IAUC for pilau rice served with curry was also significantly lower than that of pilau rice (P=0.001) (Table 3.5 and Figure 3.3).

The mean change in blood glucose concentration from baseline (Table 3.6) was compared for two pairs of test foods respectively (i. chapatti with and without curry and ii. pilau rice with and without curry) at the time points blood glucose was measured. The changes in blood glucose levels at 15, 30, 45 and 90min were significantly lower after consumption of chapatti served with curry meal compared to chapatti eaten alone (Table 3.6; Figure 3.4 and Figure 3.5). At the peak glucose rise at 45min, mean change in blood glucose for chapatti served with curry $(1.8\pm0.2 \text{ mmol/l})$ was significantly lower (*P*=0.013) than for chapatti eaten alone (2.7±0.3 mmol/l).

Pilau rice served with curry meal elicited significantly lower changes in blood glucose at 15, 30, 45 and 60min compared to pilau rice eaten alone (Table 3.6; Figure 3.6 and Figure 3.7). The glucose rise for both test meals peaked at 30min where mean change in blood glucose for pilau rice served with curry was1.3 mmol/l lower (P=0.001) than for pilau rice.

3.5.4 GI measurement values of foods/mixed meals

Table 3.7 shows the GI value and GI classification of each test food. The GI values are classified into categories considered as eliciting a low (<55), medium (55-69) or high (\geq 70) glycaemic response (Brand-Miller et al., 2003). The test foods ranged from low (chapatti served with curry, pilau rice served with curry) to medium (chapatti, rice, pilau rice) GI. The mean GI values for chapatti (68), rice (66) and pilau rice (60) did not differ significantly when tested using one way ANOVA. When tested using paired t-test, mean GI values of chapatti served with curry (45) was significantly lower (P=0.004) compared to chapatti eaten on its own (68). Pilau rice served with curry also elicited significantly lower (P=0.001) GI (41) in comparison to pilau rice (60) without curry.

	Mean	SEM	SD	Range
Age	27.9	1.3	5.1	20.0 - 34.0
Weight (kg)	53.7	2.5	9.7	41.0 - 74.2
Height (cm)	159.7	2.4	9.4	147.0 - 180.0
BMI (kg/m ²)	20.9	0.5	1.8	18.5 - 24.2
Waist circumference (cm)	70.1	1.4	5.5	64.0 - 84.0
Body fat percentage (%)	20.3	1.2	4.8	14.9 - 34.4
Fasting blood glucose (mmol/l)	4.8	0.1	0.3	4.4 - 5.3

Table 3.1: Anthropometric and clinical characteristics of subjects studied (n=15)

SEM= standard error of mean; SD= standard deviation

Table 3.2: Nutrient composition of test foods providing approximately 50g of carbohydrate

	Ingredients	Nutrition information				
		Energy (kcal)	Carbohydrate (g)	Protein (g)	Fat (g)	Fibre (g)
Chapatti	78 g Wheat flour ¹ 50 ml water	251	50.3	9.4	1.5	5.5
Chapatti served with curry	Chapatti: 73 g wheat flour ¹ 47 ml water Chicken curry ² : 25 g corn oil 63 g chicken 45 g tomatoes 20 g onion 1 g curry powder	545	50.3	23.5	28.0	6.1
Rice	65 g Basmati rice ¹ 120 ml water	234	50.0	5.8	1.0	0.0
Pilau	65 g Basmati rice ¹ 120 ml water 3 g vegetable oil ² 1 g onions ² 1 g curry powder ²	264	50.4	5.9	4.1	0.0
Pilau rice served with curry	Pilau rice: 61 g Basmati rice ¹ 113 ml water 3 g vegetable oil ² 1 g onions ² 1 g curry powder ² Chicken curry ² : 25 g corn oil 63 g chicken 45 g tomatoes 20 g onion 1 g curry powder	559	50.4	20.2	30.7	0.9

¹ Nutrient content derived from nutrition information on food packaging and ²nutrient analysis software WinDiet 2005

Table 3.3: Food preparation

Items Method of preparation*

- Chapatti To make the chapatti dough, a well was made in the centre and warm water was stirred in a little at a time to form the dough. The dough was then kneaded (5 min), placed in a bowl and covered with cling film and left to stand (15 min). Then the dough was kneaded again (5 min), rolled out into a thin disc using a rolling-pin and cooked both sides on a heated griddle at medium heat.
- Rice Raw basmati rice was put in an automatic electric rice cooker. Then water was added and cooked (13 min). After the rice was cooked, it was left to stand in the rice cooker (2 min), taken out, put on a plate and covered with cling film to avoid it from drying out.
- Pilau Rice Onions were sautéed in vegetable oil until fragrant and golden brown. Then curry powder and raw basmati rice were stirred in. This mixture was then transferred to the rice cooker and water was added. The rice was cooked (13 min) and left to stand in the rice cooker for 2 min. Then it was taken out, put on a plate and covered with cling film to avoid it from drying out.
- Curry Oil was heated in the pan. Onions were sautéed in corn oil until fragrant and golden brown. Then curry powder and tomatoes were added and stirred in. Finally, chicken was added and left to simmer on medium heat stirring occasionally (10min).

All ingredients were weighed according to the recipe as listed in Table 3.2

Test Food	Ν	¹ Mean blood glucose (mmol/l)	SEM	SD
Chapatti	10	4.7	0.1	0.4
Chapatti served with curry	10	4.8	0.1	0.4
Pilau rice	11	4.9	0.1	0.4
Pilau rice served with curry	11	4.8	0.2	0.6
Rice	10	4.6	0.1	0.2
² Reference (glucose)	15	4.8	0.1	0.3

Table 3.4: Mean fasting blood glucose (mmol/l) before test food and reference food

¹ mean of fasting blood glucose values were not significantly different when tested using One way ANOVA (p=0.635)

² mean of two fasting blood glucose measurements for each subject
Test Food	Ν	Mean IAUC	SEM	SD	<i>p</i> -value ¹
Chapatti Chapatti served with curry	10 10	183.6 110.1	7.0 12.0	22.3 38.0	0.002
Rice	10	180.6	22.3	70.4	
Pilau rice	11	162.4	16.8	55.8	0.001
Pilau rice served with curry	11	111.9	18.8	62.3	

Table 3.5: Mean incremental area under the blood glucose curves (IAUC) for each test food

SEM= standard error of mean; SD= standard deviation; All test foods were tested against glucose as reference food.

¹ the mean GI values for chapatti and chapatti served with curry (n=9) and pilau rice and pilau rice served with curry (n=9) were significantly different (p<0.05) when tested using paired comparison T-test.

Time (min)	Chapatti	Chapatti served with curry	<i>p</i> -value	Pilau rice	Pilau rice served with curry	<i>p</i> -value
15	1.1 ± 0.2	0.5 ± 0.2	0.021	1.1 ± 0.2	0.5 ± 0.2	0.009
30	2.5 ± 0.1	1.8 ± 0.2	0.030	2.8 ± 0.3	1.5 ± 0.2	0.001
45	2.7 ± 0.3	1.8 ± 0.2	0.013	2.2 ± 0.2	1.6 ± 0.2	0.007
60	2.3 ± 0.3	1.4 ± 0.2	0.065	1.9 ± 0.2	1.2 ± 0.3	0.036
90	1.0 ± 0.1	0.6 ± 0.2	0.047	1.0 ± 0.3	1.0 ± 0.2	0.781
120	0.7 ± 0.1	0.4 ± 0.2	0.229	0.7 ± 0.2	0.6 ± 0.3	0.598

Table 3.6: Mean¹ change in blood glucose (mmol/l) of healthy subjects after consumption of test foods measured at specific time points

¹Mean ± standard error of mean

Statistical significance was set at p < 0.05 for t-test (n=9)

Blood glucose readings were taken starting at time 0 min and subsequently after 15, 30, 45, 60, 90 and 120 mins.

Test Food	Ν	Mean Gl ¹	SEM	SD	GI classification	<i>p</i> -value ²
Chapatti	10	68	8	23.9	Medium	
Chapatti served with curry	10	45	5	16.5	Low	0.004
Rice	10	66	7	23.5	Medium	
Pilau rice	11	60	6	18.9	Medium	0.001
Pilau rice served with curry	11	41	5	16.2	Low	

Table 3.7: Glycaemic Index (GI) of test foods

SEM= standard error of mean; SD= standard deviation; All test foods were tested against glucose as reference food

¹ the mean GI values for chapatti (n=10), rice (n=10) and pilau rice (n=11) were not significantly different when tested using one way ANOVA.

² the mean GI values for chapatti and chapatti served with curry (n=9) and pilau rice and pilau rice served with curry (n=9) were significantly different (p<0.05) when tested using paired comparison T-test.



Figure 3.1: Blood glucose responses (Mean±SEM) in healthy subjects after consumption of chapatti (n=10), chapatti served with curry (n=10), pilau rice (n=11), pilau rice served with curry (n=11) and rice (n=10) each containing 50g of available carbohydrate. Each point represents the average blood glucose concentration for subjects at that time point. Standard errors of the mean values are represented by vertical bars.



Figure 3.2: Blood glucose responses (Mean±SEM) in healthy subjects after consumption of chapatti and chapatti served with curry each containing 50g of available carbohydrate. Each point represents the average blood glucose concentration for 10 subjects at that time point. Standard errors of the mean values are represented by vertical bars.



Figure 3.3: Blood glucose responses (Mean±SEM) in healthy subjects after consumption of pilau rice and pilau rice served with curry each containing 50g of available carbohydrate. Each point represents the average blood glucose concentration for 11 subjects at that time point.



Figure 3.4: Change in blood glucose (Mean±SEM) in healthy subjects (n=10) after consumption of chapatti. Each point represents the average change in blood glucose for 10 subjects. Standard errors of the mean values are represented by vertical bars.



Figure 3.5: Change in blood glucose (Mean±SEM) in healthy subjects (n=10) after consumption of chapatti served with curry. Each point represents the average change in blood glucose for 10 subjects. Standard errors of the mean values are represented by vertical bars.



Figure 3.6: Change in blood glucose (Mean±SEM) in healthy subjects (n=11) after consumption of pilau rice. Each point represents the average change in blood glucose for 11 subjects. Standard errors of the mean values are represented by vertical bars.



Figure 3.7: Change in blood glucose (Mean±SEM) in healthy subjects (n=11) after consumption of pilau rice and curry. Each point represents the average change in blood glucose for 11 subjects. Standard errors of the mean values are represented by vertical bars.



Figure 3.8: Change in blood glucose (Mean±SEM) in healthy subjects (n=10) after consumption of rice. Each point represents the average change in blood glucose for 10 subjects. Standard errors of the mean values are represented by vertical bars.

3.6 Discussion

Population-based studies usually derive dietary GI and GL values from food frequency questionnaires (FFQ) and this method is known to have validity issues arising from the fact that mixed-meals are assigned a GI value based on an equation (Wolever et al., 1994) which does not account for the potential glycaemic lowering effect of adding fat and/or protein in the mixed meal. Consequently the GI value assigned to a mixed meal might not be accurate. Indeed, co-ingestion of carbohydrate foods with protein and fat (Moghaddam et al., 2006) and addition of toppings to foods (Henry et al., 2006) has been shown to lower the GI of the meal/food. This was the rationale for measurement of the glycaemic response to three key South Asian staples with and without curried chicken meals. These GI values were subsequently used in the estimation of dietary GI and GL (Chapter 6).

In the current study, the addition of curry sauce and chicken to staple foods typical of South Asian diets, containing 28-30 g of fat and 20-23 g of protein per 50 g of carbohydrate, effectively reduced the glycaemic response to these foods compared to eating them alone and shifted their GI values from the medium to low category. It is important to understand the relationship between the recipe of a meal and its GI value so that better approximations can be made in epidemiology and other studies. The use of published GI values for individual foods to predict total GI of meals results in an overestimation of the total GI value of meals as demonstrated by others (Flint et al., 2004;Dodd et al., 2011). It has been established that addition of fat lowers the glycaemic response to a carbohydrate meal (MacIntosh et al., 2003;Owen and Wolever, 2003). The present study confirmed these findings.

The present study was well designed and controlled in the sense that the carbohydrate content of all meals were kept constant (50g) and the same subjects were used as a control for paired meals. The coefficient of variation for the reference food was well within the usual range observed for GI measurement studies. Therefore, any change in the glycaemic response when the staples were eaten combined with curry sauce in a traditional mixed meal was a result of changes in energy, fat, protein and fibre content in the meals.

The fat content was kept to a close range (28–39 g per 50 g carbohydrate) between both meals. Thus, the effect of fat is highly significant. The fibre content of meals was higher for chapatti-based meals compared to rice (6 g), although the total fibre contribution was very low. Furthermore, other studies have shown that the fibre content in a meal does not predict the glycaemic responses (Fabricatore et al., 2011) and variations in fibre content within the same foods do not affect the glycaemic response (Thondre et al., 2012). The role of protein in the reduction of GI and glycaemic response is also important because the curries contained a higher amount of protein compared to the staples alone (approximately 14 g more). Protein was also constant between the two mixed meals and could have contributed to the reduced glycaemic response observed in the present study. Protein has been shown to induce high insulin responses (Hatonen et al., 2011) and also adding protein to carbohydrate-based meals has been shown to reduce glycaemic responses in mixed meals as effectively as fat (Moghaddam et al., 2006). Combining protein with fat has a stronger effect on the decrease in glycaemic response. These effects may be mediated by slowing gastric emptying (Karamanlis et al., 2007). As a result of the different amounts of fat versus protein in our tested meals, it cannot be concluded which component had a more important effect on reducing the glucose responses in our results. Protein and fat have been shown to impact on the GI values for foods in other studies (Flint et al., 2004; Henry et al., 2006;Moghaddam et al., 2006;Dodd et al., 2011).

Possible mechanisms for our findings have been documented previously. Fat has been shown to reduce the glycaemic response by delaying gastric emptying and reducing glucose absorption. These mechanisms are mediated by fat-induced incretin responses such as enhancing gastric inhibitory polypeptide (Collier and O'Dea, 1983) and glucagon-like pepide-1 secretion (Thomsen et al., 1999). Furthermore, insulin responses are augmented when either fat or protein alone is added to a carbohydrate- based meal (Ercan et al., 1994;Hatonen et al., 2011). When fat and protein are part of a normal mixed meal, fat elicits an increase in insulin which is a result of incretin mediation, whereas protein does not have a similar effect (Carrel et al., 2011). The GI values of the foods in the current study were not comparable to other published GI values for similar foods because the tested meals (e.g. chapatti and curry or rice and curry) differed in ingredients and cooking methods. Chaturvedi et al. (1997) reported GI values for tomato rice curry and chapatti with curry in Pakistan using recipes that included the addition of tomato (25 g), oil (10 g) and bottle gourd (100 g), which are different from our recipes. Chew et al. (1988) investigated glycaemic responses to Indian rice served with lentils and cauliflower curry in Australia (recipe not specified) while Hettiaratchi et al. (2009) determined the GIs of Sri Lankan wheat bread served with lentil curry as well as rice with meal accompaniments including lentil curry . Hettiaratchi et al. (2011) and Pirasath et al. (2010) measured the GI of parboiled rice and different types of curry (green leaf, soya meat gravy and a combination of both); however, the exact fat content that was mainly derived from coconut milk was not reported.

The GI values for the carbohydrate staples obtained in this study differed from other studies. In this study, GI values for chapatti were higher than previously reported values (Radhika et al., 2010) (68 versus 45). The nutrient content of both chapattis was comparable, although variations in other aspects which influence starch digestibility, such as wheat variety and gluten content, flour processing and cooking methods (e.g. preparation of the chapatti dough as a result of kneading of the dough and the time left to stand before cooking) could have contributed to these differences (Sharavathy et al., 2001).

Furthermore, in this study, results with respect to the GI values for basmati rice and pilau rice are similar to those reported by Henry et al. (2005a). By contrast, much lower GI values for basmati rice have been reported (Aston et al., 2008). Differences in our findings and those by Aston et al. (2008) may be a result of recipe and cooking procedures. In the study by Aston et al. (2008), the rice was eaten with 10 g of margarine to improve palatability, which may have contributed to a reduction in glycaemia. The basmati rice and pilau rice in the present study were similar in GI values; this suggests that the addition of small amounts of oil (3 g in the pilau rice) did not significantly reduce the GI of the rice. Other factors that may affect differences in GI among studies are related to methodological aspects of GI calculation, such as the exclusion of outliers as reported by Radhika et al. (2010). Variation in GI values have been shown even in very similar foods when tested by different laboratories (Wolever et al., 2003) by an estimated SD of 9.0 units of GI, possibly as a result of a random, within subject glycaemic response that varied from day-to- day.

The present study has provided new values for GI of foods using a standard protocol for measurement of GI. Paired comparisons of staple alone and as part of a mixed meal in the same subjects were used to allow a robust comparison of effects between meals. The study lacked an arm of carbohydrate (staple) and protein (chicken alone) for further comparisons.

3.7 Conclusion

In conclusion, adding protein and fat containing curries to staple foods traditional of South Asian meals reduced glycaemic responses and changed the category for GI. GI values for meals and not single foods are needed to estimate the impact upon human diet and health. Chapter 4 Agreement between weighed food records (WFR) and food frequency questionnaires (FFQ) for carbohydrate food intake of subjects in the study on dietary glycaemic index, glycaemic load and insulin resistance (HOMA_{IR}) of South Asians in Glasgow, UK

The study on glycaemic index (GI) measurements of some key staples in the South Asian diet was presented in the previous chapter. The GI values of the foods that were measured from that study was used in the calculation of the dietary GI and glycaemic load (GL) of the study subjects in the major study on dietary GI, GL, other carbohydrate-related dietary variables and HOMA_{IR} (Chapter 6). In this chapter, a measure of assurance of whether the carbohydrate food intakes reported by study subjects reflected their habitual diet was obtained. It was imperative to ensure that the weighed food records (WFR) represented the habitual diet because one of the objectives of the study in chapter 6 was to estimate the habitual dietary GI and GL of study subjects from WFR. Therefore determination of dietary GI and GL were based on the assumption that the diets reported in WFRs were habitual.

4.1 Introduction

Both FFQs and food records are dietary assessment methods that are used to measure dietary intake in nutrition research (Thompson et al., 2010). The FFQ (Willett, 2013) is the most frequently used dietary assessment method to assess habitual diets in epidemiological studies, given that it is relatively inexpensive, easy to administer, less labour demanding when compared with, for example, food diaries or 24-h dietary recalls. The FFQ method asks respondents to report their habitual frequency of consumption of each food from a list of foods for a specific period.

Weighed food records (WFR) have been considered as reference method of dietary assessment or referred to as the "imperfect gold standard" (Coulston and Boushey, 2008). A valid (or accurate) diet record is defined as a complete and accurate record of all foods consumed on specific number of days where

everything consumed on those recorded days are foods and beverages that the subject would have consumed in usual circumstances (ie. had they not been involved in a study) (Livingstone and Black, 2003). Although the weighed food intake method is relatively accurate in that subjects directly weigh their foods instead of estimate their food intakes by portion sizes, it is also known to place an appreciable amount of burden on respondents which could lead them to alter their usual food habit and therefore, result in biasness or misreporting in the estimate of food and beverage intakes (Livingstone et al., 1990;Rebro et al., 1998;Vuckovic et al., 2000;Gibson, 2005;Poslusna et al., 2009).

In the current study, the WFR includes subjects' habitual food and beverage intake for up to 7 days (3 to 7 days for the current study) while the FFQ includes intakes for a longer period (in the past 6 months for the current study) therefore possibly capturing a more habitual diet pattern. In consideration of this and the limitations of the WFR, good agreement between the WFR and FFQ would provide some additional evidence that the study subjects' WFR (Chapter 6) reflect their habitual dietary GI and GL. Weighed food records have been found to have the least correlated errors with food frequency questionnaires (Cade et al., 2004).

Participants of this major study on dietary variables and HOMA_{IR} were asked to complete weighed food records (WFR) or food inventory for up to a week from which habitual dietary GI and GL were calculated from. Before carrying out the WFR, subjects were asked to complete a food frequency questionnaire (FFQ) to report their usual intake of carbohydrate foods in terms of frequency and amount of intake as the FFQ is a dietary assessment tool, like the WFR, that can also be used to estimate habitual dietary intakes although for a longer time amount of time than the WFR.

The FFQ used in this study has previously been validated against weighed food records in a South Asian population (Kassam-Khamis et al., 1999). Therefore, these two methods should have good agreement in nutrient intakes estimation and thus intakes of carbohydrate foods as well. Based on this assumption, the aim of the study was to obtain a measure of agreement between carbohydrate food intakes that were reported in the WFR and FFQ. This was investigated in

several ways. Firstly the ranking of carbohydrate foods (staples) based on study subjects' reported average amounts of daily consumption (g/day) from the WFR and FFQ was observed. Secondly, the agreement between WFR and FFQ were determined using correlation analyses (Bivariate Spearman Correlation). In addition to correlation analyses, Cross-classification analysis, Weighted Kappa analysis as well as Bland and Altman analysis was carried out to examine agreement between the WFR and FFQ method for carbohydrate intake.

4.2 Study Objectives and hypothesis

Objective

To determine the agreement between the WFR and FFQ method for carbohydrate intake by gender (males, females) and ethnicity (South Asian, European).

Hypothesis

There would be good agreement between the average total amount of staples in the diet reported (g/day) in the WFR and FFQ.

4.3 Subjects and Methods

4.3.1 Subjects

The study subjects in this chapter (Chapter 4), Chapter 5 and Chapter 6 were participants of the major study on dietary glycaemic index, glycaemic load, other carbohydrate-related dietary variables and HOMA_{IR} in South Asians in Glasgow, UK. The initial aim was to recruit a target sample of a total of 152 subjects of which 76 would be of South Asian ethnicity (38 males and 38 females) and 76 would be Europeans (38 males and 38 females). This was estimated to give 80% power to detect 5% statistical significance in the percentage of energy intake derived from dietary carbohydrate between ethnic groups for each respective gender with an effect size of 0.66 derived from Sevak et al. (1994). Sample size calculations were estimated using G*power 3.1 program (Faul et al., 2007). A total of 142 subjects were recruited for this study. The number of subjects who completed the whole study comprised of a total of 111 subjects of which 52 were South Asian (30 males and 22 females) and 59 were Europeans (30 males and 29 females). Study compliance is described under subtopic 4.4.3.

Recruitment and general criteria of study subjects are detailed in Chapter 2, General Methods. Ethical approval to conduct the major study was granted by the Medical Faculty Ethics Committee, University of Glasgow. Study subjects were healthy volunteers recruited from the local community in Glasgow, UK. All subjects had resided in Glasgow, UK for at least six months and of either South Asian (from Pakistan) or European ethnicity.

4.3.2 Anthropometric measurements

Anthropometric measurements such as height, weight, waist circumference and body fat percent were taken in the morning on subjects wearing light clothing and no shoes using standard methods of measurement as described in Chapter 2, General Methods, Subtopic 2.4 Anthropometric measurements.

4.3.3 Food frequency questionnaire

Because the primary population under study were South Asians, a food frequency questionnaire (FFQ) most suitable for this population was used (Kassam-Khamis et al., 1999). This FFQ was designed for a population-based case-control study of diet and breast cancer to evaluate macro and micronutrient intake among South Asian migrants residing in the UK. The FFQ had been validated against 24-hour recalls in a total of 133 South Asian women and was found to be of reasonable validity (Sevak et al., 2004). The FFQ was adapted to suit the purpose of our study as elaborated below.

The FFQ used in this study (Appendix V) was an adapted version of the FFQ by (Kassam-Khamis et al., 1999). The FFQ in this present study listed carbohydrate foods from the original FFQ categorised into breads, cereals, rice, and lentils, pulses and dahls. The adapted portion of the FFQ were addition of several carbohydrate rich food items such as Pasta, Spaghetti, Pizza, Potatoes (potatoes -in curry, potatoes -chips/fries, potatoes -baked), and Pakora which were not in the original FFQ. This is because these are also carbohydrate rich foods consumed by South Asians. A column to record the brand of the food product frequently consumed was also added. Blank rows were provided in the FFQ to allow recording of any food items that were not listed in the FFQ but frequently consumed by the subjects. The FFQ also include columns for the portion size (in grams, for a medium serving of the food), frequency of food intake in terms of how many times were eaten in a day, week, month or rarely/never.

As explained by (Kassam-Khamis et al., 1999), the standard portion size of the items listed in the FFQ were obtained from MAFF. (1994) which is based on Gregory et al. (1990). For foods with no portion size listed in the FFQ, subjects were provided with a photographic atlas of food portion sizes (Nelson et al., 1997) to refer to. Subjects chose the photo of the foods which, in their opinion, best represented their usual medium serving portion size for that particular food item.

4.3.4 Administration of the food frequency questionnaire

The FFQ was self-administered because it enabled subjects to relax and recall their frequency of food consumption without having the stress of an interviewer waiting on an answer (subjects were told to take all the time they needed). Subjects were instructed to recall and choose one column which best reflected their consumption pattern (whether the food item was usually consumed daily, weekly, monthly or rarely/never) over the past 6 months and then to write down the number of times they usually ate the food item in that column and identify the portion size usually consumed by using the Photographic Atlas of Portion Sizes (Nelson et al., 1997) as a guide, where indicated. Prior to completion of the FFQ, subjects were shown the FFQ and given verbal and written instructions to ensure that they understood how to complete it. Upon completion of the FFQ, it was examined and subjects were interviewed to fill in any blank or unclear entries.

4.3.5 Conversion of food frequency to amount of food intake

For each subject, the data from the FFQ such as the reported pattern of consumption (daily, weekly, monthly or rarely/never), frequency of intake per day (1 time, 2 times etc.) and weight of the portion size of the food item usually consumed at each time was entered into Microsoft Excel spreadsheet. The quantity (weight in grams) of a carbohydrate food item consumed in total per day was calculated using Formula 4.1 as shown below. The weight of the food item consumed per day by each subject was then entered into IBM SPSS Statistics 21 (IBM Corp., Armonk, NY) for statistical tests.

Formula 4.1

For each subject,

weight of the food item consumed in total per day =

(*Conversion Factor) (no. of times consumed per day, week or month) x weight of the portion size consumed in grams

*The Conversion factor is 1.0 if consumed daily, 0.14 if consumed weekly and 0.03 if consumed monthly

4.3.6 Weighed food records

Subjects were required to complete a food inventory or weighed food record. They were provided with a food inventory or weighed food record (Appendix VI) and a digital kitchen weighing scale with an "add and weigh function" accurate to ±1g and the capacity to weigh up to 3000grams. They were asked to record the time, place and meal (breakfast, lunch, dinner, snack) taken every day for up to seven days (Monday to Sunday). Data for subjects who recorded at least three days of food and drink intake, one being a weekend day, were deemed acceptable and included in our analysis. This is because although food intake reporting periods of longer than three days and ideally seven days would be more accurate, the mean energy intakes for foods recorded in three and seven days would still represent an average of habitual food intakes (Whybrow et al., 2007).

Subjects were given a demonstration on how to weigh foods and drinks using the scale provided and given written instructions and examples on how to record their foods and drinks in the food record. They were asked to include all food and drinks consumed over the recording days including any leftovers, all food and drink items consumed at home or prepared at home and consumed elsewhere and all items consumed outside home.

To allow improved accuracy of the food descriptions and to facilitate accurate food entry into nutrient analysis software and assignment of GI values to foods, they were asked to include brand names of products and recipes (ingredients and cooking method if known), encouraged to keep food wrappings/packages of foods, and to write as much information about them as they can in the food record. In cases where it was not possible to use weighing scales (for items which were too light to be weighed, for example a very small quantity of salt), subjects were asked to describe quantity using household measures (teaspoon, tablespoon etc.).

Subjects were asked inform the researcher if their intake was not representative of their usual eating habits (for instance if they fell sick or attended a party where they did not eat as usual). On the second day of dietary recording, the subjects were called to ensure that recording was being done. When the food dairies were returned, they were checked for completion. In cases where clarification or confirmation of a particular detail in a food record was required, the subjects were contacted either in person or over the telephone depending on the subject's convenience.

4.3.7 Analysis of weighed food records

Weighed food records were analysed with dietary analysis software, WinDiets 2005 (The Robert Gordon University, Aberdeen, UK). WinDiets includes the complete UK database as published in the 5th edition of McCance and Widdowson's Food Composition Tables and supplements. Food items from each study subject's diet dairies were entered into WinDiets to obtain average nutrient intake (namely carbohydrate, protein, fat and fibre).

If a food item(s) recorded in the food dairy was not found in WinDiets, the nutrient composition of the food was obtained from the most appropriate and available resource such as nutrition information on the food packages/wrappers and published food tables from Kassam-Khamis et al. (2000) which were entered manually into WinDiets 2005.

4.3.8 Relative validation of weighed food records

Subjects' weighed food records were examined for relative validity using the Goldberg cut-off method (Goldberg et al., 1991) in line with the guidelines for its calculation and use (Black, 2000). The Goldberg cut-off method was used because it is an established method, not invasive and less expensive compared to other methods of validating self-reported energy intakes such as the use of biomarkers (urinary nitrogen) and doubly labelled water.

Using the Goldberg cut-off method, likely underreporters of energy intake was identified as having a ratio of reported energy intake to predicted BMR (or EI_{rep} :BMR) that fell below the 95% Confidence Limit (CL) of the subject's physical activity level (PAL_{rep}) which was derived from self-reported physical activity records. Therefore each subject had their own individual PAL cut-off value at the lower 95% CL. This cut-off was calculated using the Goldberg cut-off equation (Black, 2000). EI_{rep} was estimated from subjects' weighed food

record (see method in subtopic 4.3.6). Subjects' BMR was predicted from Oxford BMR equations (Henry, 2005). PAL_{rep} was derived from subjects' self-reported physical activity record (see method in subtopic 5.3.3). By this individual cutoff approach, the percentage of possible underreporters from the total number of subjects (n=111) was 68% and the percentage of possible underreporters by gender and ethnicity is as follows: South Asian males, 73%; European males, 70%; South Asian females, 59% and European females, 66%. However, all subjects had El_{rep}:BMR that was equal to or higher than 1.2. When the cut-off was set at lower 99% CL of PAL_{rep}, the El_{rep}:BMR of all subjects were well above the cut-off which deemed them to be acceptable reporters of energy intake.

4.3.9 Determination of habitual carbohydrate food intake (types and amounts) from weighed food records

A list of carbohydrate food items, similar to the carbohydrate food items in the FFQ, was entered into excel spreadsheet. Then the quantity (grams) of each carbohydrate food reported to be consumed (from the weighed food record) by each study subject, was entered manually in that excel spreadsheet for every day that food intake was recorded. Average daily consumption (grams/day) of the carbohydrate food was then calculated separately by gender and ethnicity.

The types of foods that were mostly eaten in combination with main staples (rice, chapatti, potatoes, pasta) were identified from the weighed food record of each subject and entered into excel spreadsheet. Then the quantity (grams) of the foods reported to be consumed with main staples (from the weighed food record) by each study subject, was entered manually in that excel spreadsheet for every day that food intake was recorded. Average daily consumption (grams/day) of foods that were eaten with main staples was then calculated separately by gender and ethnicity.

4.3.10 Statistical Analysis

Statistical analyses were performed with IBM SPSS Statistics 21 (IBM Corp., Armonk, NY). Descriptive statistics of the study participants' characteristics were computed and presented. All variables for which statistical analyses were carried out on were examined for normality (Shapiro Wilks statistics test for normality) and outliers (Box-plot or box-and-whisker plots). Means and standard deviation for variables were presented.

There is currently best statistical method recommended in the literature for assessing the validity of dietary assessment tools (Masson et al., 2003). Therefore several statistical methods of assessing relative agreement between FFQ and WFR were used. These included correlation coefficients (Spearman correlation tests was used as most data were not normally distributed), crossclassification, weighted kappa statistics and Bland and Altman statistics.

In the cross-classification method, subjects were categorised into thirds of intake by the FFQ and WFR and then the percentage of subjects correctly classified into the same category and grossly misclassified into the opposite category was calculated.

For weighted kappa statistics (K_w) Values of kappa above 0.80 indicate very good agreement, between 0.61 and 0.80 good agreement, 0.41-0.60 moderate agreement, 0.21-0.40 fair agreement and 0.20 poor agreement (Altman, 1991) between FFQ and WFR. Relative agreement between methods was also assessed using Bland and Altman statistics between FFQ and WFR (Altman, 1991).

4.4 Results

4.4.1 Average amount of carbohydrate foods (grams/day) estimated from weighed food records and food frequency questionnaires

Spearman Correlation

4.4.1.1 South Asian males

Table 4.1 shows the average amount of carbohydrate foods consumed by South Asian males estimated from weighed food records (WFR) and food frequency questionnaires (FFQ) respectively. Carbohydrate staples reported to be consumed ranked from highest to lowest average consumption per day according to WFR were unleavened bread (163.12g) followed by rice (90.65g), bread (white, wholemeal and brown bread) (62.64g), potatoes (32.41g), pasta (25.07g) and breakfast cereals (16.83g). The FFQ ranked these staples similarly (the mean of unleavened bread and rice estimated from the FFQ were not markedly different) with the exception of potatoes which were reported, through the FFQ, to be consumed more than bread.

For types of unleavened bread, the WFR ranked mean daily consumption of, Chapatti/Roti (without fat) as highest followed by Naan/Pitta bread, Paratha and Chapatti/Roti (with fat). In the FFQ, reported amounts of these foods except for Chapatti/Roti (with fat) were almost equal. The WFR and FFQ both showed that Chapatti/Roti (with fat) was the type of unleavened bread that was least consumed by South Asian males.

Boiled white rice (Basmati, long grain white rice) was consumed more than Pilau/Biryani rice according to the WFR but this was the other way around based on the FFQ. The standard deviation for the mean of Pilau/Biryani estimated from the FFQ was relatively high at 145.66g. South Asian males reported that they consumed more white bread/rolls compared to wholemeal bread/roll as shown in both the WFR (55.24g and 7.40g, respectively) and FFQ (24.36g and 10.02g, respectively). High fibre and low fibre breakfast cereals were observed to be consumed in relatively lower amounts compared to other staples and this ranged from 3.37g to 10.23g in WFR and FFQ combined.

4.4.1.2 European males

In European males, carbohydrate staples reported to be consumed ranked from highest to lowest consumption according to WFR was bread (white, wholemeal, brown) followed by pasta, potatoes, rice and breakfast cereals (Table 4.2) ranging from as low as 6.44g/day for unleavened bread for instance, to as high as 115.36g/day for bread (White, Wholemeal, Brown). Based on the FFQ, the ranks for these foods with the exception of breakfast cereals, were not markedly different as mean intakes were within similar ranges (59.93 for rice to 88.71g for potatoes).

According to WFR, mean consumption of white bread/roll (92.17g/day) was higher than wholemeal bread/roll (23.30g/day) but based on the FFQ, mean consumption for these foods were quite similar where wholemeal bread/roll was reported to be consumed only 9.43g/day more than white bread/roll. For all other staples, the WFR and FFQ ranks were similar where boiled white rice (43.01g/day and 51.45g/day, respectively) was reported to be consumed noticeably more than rice cooked with oil like Pilau/Biryani (3.40g/day and 8.48g/day, respectively). Both the WFR and FFQ similarly showed that high fibre breakfast cereals were consumed more than low fibre, sugared varieties and Naan/Pitta bread was observed to be the type of unleavened bread mostly consumed by European males (5.66 g/day estimated from WFR; 11.78g/day estimated from FFQ).

4.4.1.3 South Asian females

Reported average daily consumption of staples in the diet for South Asian females estimated from the WFR and FFQ were similar (Table 4.3). Both dietary methods yielded mean consumptions which was noticeably the lowest for breakfast cereals (19.63 g/day from WFR and 16.92g/day from FFQ). If ranked according to absolute mean amounts by WFR, unleavened bread ranked highest followed by rice, bread, potatoes and pasta but considering SD values, the mean consumption of these staples were actually quite similar ranging from, for instance, 50.03g/day (SD of 65.01) for pasta to 84.71g/day (SD of 80.58) for unleavened bread.

In contrast to this, the FFQ ranked the mean consumption of rice to be the highest (141.99g/day) among the staples in the diet. However, in agreement with the WFR, the FFQ showed that the reported mean consumption of other staples such as unleavened bread, bread, potatoes and pasta were quite similar. Mean consumption of unleavened bread (67.23g/day) for instance, was not markedly different from intakes of potatoes (54.64g/day) whereas intakes of pasta (41.05g/day) were not that different from bread (37.39g/day).

4.4.1.4 European females

For European females, carbohydrate staples reported to be consumed ranked from highest to lowest mean consumption per day according to WFR, was pasta (79.98g) followed by bread (63.85g), potatoes (51.07g), rice (33.75g) and breakfast cereals (5.35g) (Table 4.4). These ranks were similar to estimates from the FFQ with the exception that in the FFQ, reported consumption of potatoes was ranked higher than bread intakes but only by 10.54g higher. The WFR showed similar mean intakes of wholemeal bread/roll and white bread roll differing only by 8.14g/day but in the FFQ, consumption estimates for wholemeal breads/rolls (46.26g/day) were higher than for white bread/rolls (13.93g/day).

Average consumption estimates from both the WFR and FFQ for boiled white rice (31.94g/day and 23.30g/day, respectively) was higher than rice cooked with

oil (Pilau/Biryani) (1.48g/day and 15.25g/day, respectively). For breakfast cereals, consumption estimates from the WFR and FFQ were also observed to be similar where high fibre varieties (23.12g/day and 17.01g/day, respectively) were consumed more than low fibre/sugared types (4.28g/day and 8.72g/day, respectively). The WFR and FFQ also agreed that unleavened breads in total were the least consumed staples in the diet and that Naan/pitta breads were the type of unleavened bread mostly consumed by European females. 4.4.2 Agreement/correlation between weighed food records and food frequency questionnaires in carbohydrate intakes

4.4.2.1 South Asian males

In South Asian males, the average daily consumption amounts of all types of carbohydrate foods/staples estimated from WFR were found to relate positively and significantly to estimates from the FFQ (Table 4.5). The relationship was strong for breakfast cereals (r=0.723), unleavened bread (r=0.719) and pasta (r=0.666) but moderate for rice (r=0.484), bread (r=0.467) and potatoes (r=0.424).

4.4.2.2 European males

In Table 4.6, European males' WFR estimates were observed to relate positively to estimates from their FFQ for average daily consumption of four out of six staples namely potatoes, pasta, breakfast cereals and rice was moderately strong overall (r of 0.554, 0.470, 0.457 and 0.431, respectively) and statistically significant (P<0.05). Estimated average amounts of bread (white, wholemeal, brown) and unleavened bread respectively from the WFR was weakly positively related to estimates from the FFQ almost reaching statistical significance for bread (r=0.347, p-value=0.06) and not statistically significant for unleavened bread (r=0.025, p-value=0.894).

4.4.2.3 South Asian females

Table 4.7 shows that South Asian females were found to vary markedly in their WFR and FFQ correlations for carbohydrate foods. The correlations ranged from almost null (r=-0.002 for potatoes) to weak (r=0.339 for unleavened bread). The WFR and FFQ for four out of six staples were positively related (unleavened bread, rice, pasta and breakfast cereals) and this was statistically significant only for pasta (r= 0.527; *p*-value= 0.012). Very weak/almost null inverse relationship was observed between mean consumption ranks estimated from WFR and FFQ for bread (white, wholemeal, brown) (r= -0.069) and potatoes (r= -0.002) but this was not statistically significant.

4.4.2.4 European females

For European females, mean consumption ranks for all staples estimated from WFR and FFQ were positively related ranging from as weak as a correlation coefficient of r=0.091 for rice to as strong as r=0.674 for potatoes (Table 4.8). Relationship of moderate strength were observed for bread (r=0.589) and breakfast cereals (r=0.465). Mean consumption ranks for pasta, unleavened bread and rice that were estimated from WFR (r of 0.171, 0.238 and 0.091, respectively) were weakly related to estimates from the FFQ.

Cross-Classification, Weighted Kappa and Bland and Altman statistics

South Asian males

Kappa statistics (K_w) indicated moderate agreement between FFQ and WFR for unleavened bread and bread. Potatoes and breakfast cereal indicated fair agreement while agreement for rice and pasta were poor. Over 50% of South Asian males were correctly classified into the same third of intake for all CHO foods with the exception of rice and pasta (Table 4.5). Most subjects were correctly classified into the same third of intake for unleavened bread, followed by bread, breakfast cereals, potatoes, pasta and rice. For rice bread and potatoes, 13% of South Asian males were grossly misclassified into the opposite third of intake while only 3% or less were misclassified into the opposite third of intake for unleavened bread, pasta and breakfast cereals.

Bland and Altman statistics showed mean% of agreement for FFQ and WFR for all foods were all above 50% with the widest 95%CI and limits of agreement for potatoes with mean% agreement over 100%. Based on Bland and Altman statistics, P-value for one sample T-test, there was agreement (p-value was not statistically significant) between FFQ and WFR for 3 out of 6 CHO foods. There was agreement in intakes of rice, pasta and breakfast cereals. Intakes of unleavened bread, bread and potatoes statistically differed significantly in the FFQ and WFR.

European males

Kappa statistics (K_w) indicated fair agreement between FFQ and WFR for potatoes, pasta and breakfast cereals for which above 50% of subjects were correctly classified into the same third of intakes for these foods. Unleavened bread, rice and bread indicated only poor agreement and less than 50% of European males were classified correctly into the same third of intake for foods in these food groups (Table 4.6). Approximately 13% percent or less of European males were misclassified into the opposite third of intake for all CHO foods.

Bland-Altman statistics showed mean% agreement between FFQ and WFR to be above 100% for rice, bread, potatoes and breakfast cereal (the highest was for potatoes and the least, breakfast cereal). Unleavened bread had the lowest %mean agreement (28%). Based on Bland and Altman statistics, P-value for one sample T-test, there was agreement (p-value was not statistically significant) between FFQ and WFR for 4 out of 6 CHO foods. There was agreement in intakes of unleavened bread, rice, pasta and breakfast cereals. Intakes of bread and potatoes statistically differed significantly in the FFQ and WFR.

South Asian females

Pasta was the only CHO food that indicated moderate agreement between FFQ and WFR (K_w was 0.455). Unleavened bread and rice had fair agreement (similar K_w of 0.246) while there was poor agreement between FFQ and WFR for all other

CHO foods (bread, potatoes and breakfast cereals) (Table 4.7). At least 50% of South Asian females were correctly classified into the same third of intake for unleavened bread, rice and pasta. The same percentages (23%) of South Asian females were correctly classified into the same third and misclassified into the opposite third of intake for bread. The least percentage of South Asian females were misclassified into the opposite third of intake for bread. The least percentage of South Asian females were misclassified into the opposite third of intake for pasta (5%) and unleavened bread (9%).

Bland-Altman statistics showed mean% agreement between FFQ and WFR to be above 100% for rice, potatoes, pasta and breakfast cereal (the highest was for rice and the least was for breakfast cereal which was close to 100%). Bread and unleavened bread had %mean agreement that was 88% and below. Based on Bland and Altman statistics, P-value for one sample T-test, there was agreement (p-value was not statistically significant) between FFQ and WFR for 4 out of 6 CHO foods. There was agreement in intakes of unleavened bread, potatoes, pasta and breakfast cereals. Intakes of bread and rice statistically differed significantly in the FFQ and WFR.

European females

Kappa statistics (K_w) indicated fair agreement between FFQ and WFR for bread and potatoes while all other foods had poor agreement (Table 4.8). About 7% or less of European females were misclassified into the opposite third of intake for 3 out of 5 CHO foods (potatoes, bread and unleavened bread) while a higher % of subjects (approximately 21-28%) were misclassified into the opposite third of intake for rice, pasta and breakfast cereals .

Bland-Altman statistics showed a wide range of mean% agreement between FFQ and WFR for CHO foods; a range of 47% (unleavened bread) to 184% (potatoes). Based on Bland and Altman statistics, P-value for one sample T-test, there was agreement (p-value was not statistically significant) between FFQ and WFR for 4 out of 6 CHO foods. There was agreement in intakes of unleavened bread, bread, rice and breakfast cereals. Intakes of potatoes and pasta statistically differed significantly in the FFQ and WFR.

4.4.3 Study compliance

A total of 142 subjects were initially recruited for this study. Of this total, 111 (78%) subjects completed the study and provided complete data that were deemed valid and further analyzed. They comprised of 60 males (30 South Asians and 30 Europeans) and 51 females (22 South Asians and 29 Europeans). The total study drop-out rate was 22% (31 out of 142). Drop-out rate was higher among females (27%) than males (17%). By ethnicity, drop-out out rate for South Asians (25%) was higher than for Europeans (19%). The drop-out rate for South Asian females included those who were withdrawn from the study because it was later known that they no longer fulfilled study criteria (pregnancy). Subjects who dropped out or were withdrawn from the study did not provide complete data for analysis and were excluded from further data analysis.

The number of days weighed food intake records and Food Frequency Questionnaire were collected and the percentage of subjects who completed those records in the given days are as follows: South Asian males (n=30), 5-7 days, 77%; 3-4 days, 23% European males (n=30), 5-7 days, 87%; 3-4 days, 13% South Asian females (n=22), 5-7 days, 68%; 3-4 days, 32% European females (n=29), 5-7 days, 76%; 3-4 days, 24% Table 4.1: Average amount of carbohydrate food (g/day) intakes reported by South Asian males in weighed food intake records and food frequency questionnaires

	South Asian Males (<i>n</i> 30)						
Carbohydrate Foods	Weighe reco Mean	ed food ord SD	Food frequency questionnaire Mean SD				
Unleavened bread (all types)	163.12	124.20	90.23	106.29			
Rice (all types)	90.65	97.81	104.15	189.04			
Bread (White, Wholemeal, Brown)	62.64	43.86	34.39	40.17			
Potatoes	32.41	40.71	68.94	78.69			
Pasta	25.07	43.92	19.70	24.46			
Breakfast Cereals (High Fibre, Low Fibre, sugared)	16.83	38.12	12.17	12.71			
Chapatti/Roti (without fat)	93.65	98.69	26.41	54.95			
Chapatti/Roti (with fat)	1.78	9.74	7.70	13.49			
Naan/Pitta Bread	43.95	69.55	29.74	57.90			
Paratha	23.74	39.34	26.38	60.06			
Rice (cooked without oil, boiled)	52.47	90.19	27.20	65.92			
Rice (cooked with oil; Pilau/Biryani)	38.18	46.40	76.94	145.66			
White Bread/Roll	55.24	41.39	24.36	35.22			
Wholemeal Bread/Roll	7.40	16.18	10.02	21.53			
Breakfast Cereals (High Fibre)	10.23	36.79	3.37	5.06			
Breakfast Cereals (Low Fibre, Sugared)	6.60	12.51	8.80	10.62			

Table 4.2: Average amount of carbohydrate food (g/day) intakes reported by European males in weighed food intake records and food frequency questionnaires

	European Males (n 30)							
Carbohydrate Foods	Weighed recor Mean	l food d SD	Food frequency questionnaire Mean SD					
Bread (White, Wholemeal, Brown)	115.36	77.60	86.21	59.09				
Pasta	78.49	73.85	63.36	38.20				
Potatoes	58.95	63.00	88.71	81.27				
Rice (all types)	46.41	51.61	59.93	57.71				
Breakfast Cereals	43.96	58.20	30.73	33.31				
(High Fibre, Low Fibre, sugared) Unleavened bread (all types)	6.44	14.27	12.32	24.13				
White Bread/Roll	92.17	83.39	38.39	55.96				
Wholemeal Bread/Roll	23.20	34.70	47.82	57.90				
Rice (cooked without oil, boiled)	43.01	50.99	51.45	54.07				
Rice (cooked with oil; Pilau/Biryani)	3.40	12.97	8.48	19.86				
Breakfast Cereals (High Fibre)	35.28	57.35	24.32	32.49				
Breakfast Cereals (Low Fibre, Sugared)	8.68	18.15	6.41	8.89				
Naan/Pitta Bread	5.66	13.95	11.78	23.89				
Paratha	0.78	4.26	0.16	0.85				
Chapatti/Roti (without fat)	0.00	0.00	0.32	1.46				
Chapatti/Roti (with fat)	0.00	0.00	0.06	0.34				

Table 4.3: Average amount of carbohydrate food (g/day) intakes reported by South Asian females in weighed food intake records and food frequency questionnaires

	South Asian Females (n 22)						
Carbohydrate Foods	Weighed recor Mean	food d SD	Food frequency questionnaire Mean SD				
Unleavened bread (all types)	84.71	80.58	67.23	69.22			
Rice (all types)	74.60	57.12	141.99	101.36			
Bread (White, Wholemeal, Brown)	70.37	38.82	37.39	25.67			
Potatoes	51.11	49.19	54.64	39.13			
Pasta	50.03	65.01	41.05	41.74			
Breakfast Cereals (High Fibre, Low Fibre, sugared)	19.63	22.14	16.92	16.12			
Chapatti/Roti (without fat)	61.41	70.06	27.21	30.30			
Chapatti/Roti (with fat)	3.24	15.19	22.78	50.75			
Naan/Pitta Bread	10.17	16.06	5.81	9.38			
Paratha	9.90	24.52	11.43	29.48			
Rice (cooked with oil; Pilau/Biryani)	38.02	46.55	122.68	103.11			
Rice (cooked without oil, boiled)	36.58	34.88	19.30	24.19			
White Bread/Roll	60.10	39.50	25.07	26.73			
Wholemeal Bread/Roll	10.27	21.31	12.32	22.23			
Breakfast Cereals (Low Fibre, Sugared)	9.82	13.58	11.72	14.75			
Breakfast Cereals (High Fibre)	9.59	16.73	5.20	11.47			

Table 4.4: Average amount of carbohydrate food (g/day) intakes reported by European females in weighed food intake records and food frequency questionnaires

	European Females (n 29)							
Carbohydrate Foods	Weighed recor Mean	food d SD	Food frequency questionnaire Mean SD					
Pasta	79.98	78.60	82.54	106.75				
Bread (White, Wholemeal, Brown)	63.85	49.13	60.20	50.95				
Potatoes	51.07	38.10	70.74	70.08				
Rice (all types)	33.75	44.41	38.55	58.55				
Breakfast Cereals	27.40	39.76	25.74	29.37				
(High Fibre, Low Fibre, sugared) Unleavened bread (all types)	5.35	14.42	8.25	12.40				
Wholemeal Bread/Roll	27.85	32.39	46.26	42.95				
White Bread/Roll	35.99	36.50	13.93	39.86				
Rice (cooked without oil, boiled)	31.94	44.68	23.30	26.47				
Rice (cooked with oil; Pilau/Biryani)	1.48	7.96	15.25	45.29				
Breakfast Cereals (High Fibre)	23.12	39.27	17.01	25.76				
Breakfast Cereals (Low Fibre, Sugared)	4.28	7.97	8.72	11.27				
Naan/Pitta Bread	5.35	14.42	8.10	12.42				
Paratha	0.00	0.00	0.00	0.00				
Chapatti/Roti (without fat)	0.00	0.00	0.00	0.00				
Chapatti/Roti (with fat)	0.00	0.00	0.15	0.46				

Table 4.5: Statistics comparing carbohydrate food consumption (g/day) estimated from weighed food records (WFR) and food frequency questionnaires (FFQ) for South Asian males

	Correlation			Percentage		Bland-Altman statistics					
Carbohydrate Foods		Spearman <i>r</i>	p-value	same third	opposite third	K _w	¹ p-value for one sample T-test	² Mean % agreement	95	%CI	Limits of agreement (%)
Unleavened bread	WFR FFQ	0.719	0.000	73	0	0.600	0.001	64	36,	91	78-206
Rice	WFR FFQ	0.484	0.007	40	13	0.100	0.534	89	58,	120	58-236
Bread (White, Wholemeal, Brown)	WFR	0.467	0.009	67	13	0.500	0.004	86	28,	143	199-371
Potatoes	WFR FFQ	0.424	0.020	57	13	0.350	0.012	177	102,	253	111-465
Pasta	WFR FFQ	0.666	0.000	47	0	0.200	0.333	61	31,	91	20-143
Breakfast Cereals	WFR FFQ	0.723	0.000	60	3	0.393	0.441	97	51,	143	51-245

WFR, weighed food intake record; FFQ, food frequency questionnaire; Kw, weighted kappa ^{1}p -value for one sample T-test for testing the mean difference between WFR and FFQ with the value 0. 2 Mean % agreement =FFQ/WFR (%); 95% CI=95% CI of the mean % agreement; statistical significance was set at *p*<0.05

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Table 4.6: Statistics comparing carbohydrate food consumption (g/day) estimated from weighed food records (WFR) and food frequency questionnaires (FFQ) for European males

	Correlation			Percentage		Bland-Altman statistics					
Carbohydrate Foods		Spearman r	p-value	same third	opposite third	K _w	¹ p-value for one sample T-test	² Mean % agreement	95	%CI	Limits of agreement (%)
Unleavened bread	WFR FFQ	0.025	0.894	40	3	0.000	0.279	28	-6,	61	35-90
Rice	WFR FFQ	0.431	0.017	40	7	0.100	0.144	157	88,	226	140-454
Bread (White, Wholemeal, Brown)	WFR FFQ	0.347	0.060	47	13	0.200	0.068	128	75,	181	146-401
Potatoes	WFR FFQ	0.554	0.001	57	10	0.350	0.037	187	113,	261	165-539
Pasta	WFR FFQ	0.470	0.009	53	7	0.300	0.240	87	63,	111	18-192
Breakfast Cereals	WFR FFQ	0.457	0.011	50	3	0.250	0.265	117	58,	177	140-374

WFR, weighed food intake record; FFQ, food frequency questionnaire; Kw, weighted kappa ^{1}p -value for one sample T-test for testing the mean difference between WFR and FFQ with the value 0. 2 Mean % agreement =FFQ/WFR (%); 95% CI=95% CI of the mean % agreement; statistical significance was set at p<0.05
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Table 4.7: Statistics comparing carbohydrate food consumption (g/day) estimated from weighed food records (WFR) and food frequency questionnaires (FFQ) for South Asian females

		Correlation	n	Perc	entage jed in (%)		Bland-Altman statistics				
Carbohydrate Foods		Spearman r	p-value	same third	opposite third	K _w	¹ p-value for one sample T-test	² Mean % agreement	95	%CI	Limits of agreement (%)
Unleavened bread	WFR FFQ	0.339	0.122	50	9	0.246	0.261	88	55,	122	53-230
Rice	WFR FFQ	0.130	0.564	50	18	0.246	0.006	282	165,	398	220-783
Bread (White, Wholemeal, Brown)	WFR	-0.069	0.759	23	23	-0.161	0.005	64	34,	94	61-190
	FFQ										
Potatoes	WFR FFQ	-0.002	0.994	36	18	0.043	0.812	116	49,	181	119-351
Pasta	WFR FFQ	0.527	0.012	64	5	0.455	0.119	140	10,	290	369-649
Breakfast Cereals	WFR FFQ	0.014	0.950	36	23	0.046	0.652	104	18,	190	188-397

WFR, weighed food intake record; FFQ, food frequency questionnaire; Kw, weighted kappa ¹*p*-value for one sample T-test for testing the mean difference between WFR and FFQ with the value 0. ²Mean % agreement =FFQ/WFR (%); 95% CI=95% CI of the mean % agreement; statistical significance was set at *p*<0.05

Table 4.8: Statistics comparing carbohydrate food consumption (g/day) estimated from weighed food records (WFR) and food frequency questionnaires (FFQ) for European females

		Correlatio	n	Perc classif	centage Tied in (%)			Bland-Altm	an stati	stics	
Carbohydrate Foods		Spearman <i>r</i>	p-value	same third	opposite third	K _w	¹ p-value for one sample T-test	² Mean % agreement	95	%CI	Limits of agreement (%)
Unleavened bread	WFR FFQ	0.238	0.215	34	3	0.128	0.414	47	-11,	104	44-137
Rice	WFR FFQ	0.091	0.638	41	28	0.127	0.713	65	35,	96	42-173
Bread (White, Wholemeal, Brown)	WFR FFQ	0.589	0.001	55	3	0.327	0.703	110	68,	151	83-302
Potatoes	WFR FFQ	0.674	0.000	59	7	0.379	0.067	184	95,	273	249-617
Pasta	WFR FFQ	0.171	0.375	38	21	0.068	0.003	97	63,	130	56-249
Breakfast Cereals	WFR FFQ	0.465	0.011	41	24	0.120	0.838	131	63,	200	156-418

WFR, weighed food intake record; FFQ, food frequency questionnaire; Kw, weighted kappa ^{1}p -value for one sample T-test for testing the mean difference between WFR and FFQ with the value 0. 2 Mean % agreement =FFQ/WFR (%); 95% CI=95% CI of the mean % agreement; statistical significance was set at p<0.05

4.5 Discussion

This study aimed to determine the extent of agreement between foods and drinks reported by study subjects in WFR and FFQ. Subjects reported their food and beverage intakes in a weighed food record for 3 to 7 days. Prior to completing the WFR, subjects also reported their food and beverage consumption in an FFQ which included intakes for a longer period (in the past 6 months) therefore the FFQ possibly reflected a more habitual diet pattern. It was expected that good agreement between the WFR and FFQ would provide evidence that the study subjects' WFR (Chapter 6) reflect their habitual dietary GI and GL. WFR have been found to have the least correlated errors with FFQ (Cade et al., 2004).

Study findings indicated that overall, the WFR and FFQ mostly agreed in reported intakes of most CHO foods as observed by similar rankings of most foods. Interestingly, males compared to females, showed more agreement between the FFQ and WFR in carbohydrate (CHO) food intakes as observed from the positive and significant relationship (Spearman correlation) between the two methods for all CHO foods. Similarly kappa statistics indicated that males compared to females, showed more agreement between the FFQ and WFR in CHO food intakes although this was less pronounced through kappa statics.

Weighted kappa statistics (K_w) indicated poor to moderate agreement between WFR and FFQ for CHO food intake. There was more agreement in reporting unleavened bread, bread and pasta in the WFR and FFQ but there was particularly more variability in reporting rice, potatoes and breakfast cereals as indicated by only poor to fair agreement between WFR and FFQ for these foods.

In cross-classification analysis, more females than males were misclassified into the opposite third of intakes for CHO foods. In females, a higher percentage of South Asians than Europeans were misclassified into the opposite third of intakes for CHO foods while misclassified South Asian and European males were about equal. The percentage of subjects who were grossly misclassified into the opposite third of intakes for CHO foods was relatively low, ranging from 0% to 28%. Study subjects had wide variability (SD) in carbohydrate food intakes and the quantities of CHO reported in the WFR and FFQ varied considerably for some CHO foods like rice and pasta in South Asian females as depicted through the wide limits of agreement by the Bland-Altman statistics. However, correlation analysis and weighted kappa statistics compared to Bland-Altman statistics are more appropriate for analysis of agreement between FFQ and WFR methods (Masson et al., 2003) because Bland-Altman statistics is more applicable where absolute amounts of food intakes are concerned. The FFQ in the current study could be expected to be less accurate than the WFR in quantities of CHO food reported and the FFQ is used to rank individuals into categories/levels of intakes rather than by absolute amounts of intakes (Willett, 2013).

In female subjects, the almost nonexistent or weak correlation/agreement between the FFQ and WFR for some CHO foods may be due to the wider variety of their habitual diet. It is possible that their intakes of foods such as unleavened bread and rice in South Asian females for instance, and rice and pasta in European females, were more varied in terms of types of CHO foods eaten, how frequently it was eaten (daily, weekly, monthly or seldom) and quantities consumed which may have fluctuated from day to day. As a result, it may have been difficult for some female subjects to describe usual intakes in the FFQ for that particular food in terms of quantity and frequency of intake and this perhaps was recorded differently in the WFR, hence leading to weak correlations/less agreement between WFR and FFQ.

Rice, for instance, was reported by the South Asian females in much higher amounts in the FFQ than in the WFR particularly Pilau and Biryani. The FFQ lists Pilau (meat, vegetable) and Biryani separately so perhaps they found it difficult to estimate the frequency of their intakes which possibly varied from week to week depending on which other staples were also eaten and this led to overestimation of intakes in the FFQ. Their recorded rice intakes in the WFR should be more accurate as quantities of food and beverage intakes were measured with a weighing scale and the types of CHO recorded in the WFR considered all the different types of staples consumed by subjects in the days that they recorded their diet. Nevertheless, females also showed a good degree of consistency in reporting CHO food intakes as their WFR and FFQ were still comparable in ranking all staples except for rice. Average amounts that South Asian females reported in the FFQ and WFR at least agreed on the types of CHO foods consumed(unleavened bread, bread, potatoes, pasta and breakfast cereals) ranked from highest to lowest consumption.

Also, FFQ, compared to the WFR, puts more respondent burden in recalling information (study subjects were asked to recall their habitual diet in the past 6 months during the completion of the FFQ) and it may be more difficult for respondents to describe/estimate food portion size (Frobisher and Maxwell, 2003;Willett, 2013) as in an FFQ. However, in the current study, the use of photographic atlas of food portion size with the FFQ most likely helped respondents to recall more accurate quantities of foods and beverages reported as use of visual aids have been found to facilitate in recalling amounts of foods and beverages (Godwin et al., 2004;Subar et al., 2010) in dietary recalls.

There is also a possibility that nutrient intakes estimated from the FFQ in fact, did not agree with the estimates from WFR even though FFQ has been validated in a previous study (Kassam-Khamis et al., 1999). This is highly unlikely, however, as both the WFR and FFQ ranked almost all CHO foods for males and females similarly or at least in similar pattern and the WFR and FFQ were positively significantly related for a majority of carbohydrate foods especially in males. Other studies have also found that foods reported in FFQ and weighed food records agreed moderately (Carlsen et al., 2010;Collins et al., 2014;Fallaize and Forster, 2014).

There are several strengths and limitations on the use of the FFQ and the WFR method to obtain information on habitual diet. The FFQ is usually used in epidemiological studies to rank individuals according to their intake level rather than for measuring the absolute level of intake, therefore calculating correlation coefficients and weighted k statistics is appropriate (Masson et al., 2003;Willett, 2005). Compared to the WFR, FFQs is less burdensome for respondents, can be self-administered and quicker to complete and analyse (Gibson, 2005).

The strength of the weighed food record is that it is often considered to be more accurate compared to FFQ as respondents are asked to directly weigh foods/beverages (Coulston and Boushey, 2008). It is open-ended meaning that subjects had the freedom to express and record what they consumed. It is also widely applied in research so it facilitates comparisons to be made between studies. The WFR is, however, not without its limitations (Whybrow et al., 2015). It is relatively labour-intensive both for the respondent who has to weigh everything eaten over the days that food intake is recorded and for the researcher as well who has to analyse the food records. Use of this method requires literacy skills and everyone needs to stay motivated from beginning to the end of the study. The respondent could potentially alter their usual intakes due to inconvenience and fatigue. They can become more selective of the foods they eat so it would be easy to weigh, they may also omit or include certain foods that they do not usually consume due to their perceptions or belief of what is healthy or not in order to perhaps please the researcher.

4.6 Conclusion

In conclusion, there was better agreement (ranging mostly fair to moderate) between WFR and FFQ for carbohydrate food intakes in males than females. In males, the WFR and FFQ of Europeans and South Asians mostly similarly agreed in food intake. In females, Europeans compared to South Asians, showed slightly better agreement between the two methods in carbohydrate food intakes. The WFR and FFQ of subjects mostly agreed on unleavened bread, pasta and potatoes as reflected by the least percentage of subjects misclassified into the opposite third of intakes of these foods while there was less agreement for bread, breakfast cereal and rice. This provides evidence that the WFR possibly reflects the habitual diet. Chapter 5 Physical activity of subjects for the study on dietary glycaemic index, glycaemic load and insulin resistance (HOMA_{IR}) of South Asians in Glasgow, UK

5.1 Introduction

The risk of developing type 2 diabetes mellitus is especially high among both native and migrant South Asians (Bakker et al., 2013) and South Asians are known to have higher fasting insulin concentrations compared to white Europeans (McKeigue et al., 1988;McKeigue et al., 1991). Furthermore, South Asians having the metabolic syndrome have showed higher diastolic blood pressure, plasma triglycerides, fasting insulin and lower high-density lipoprotein-cholesterol (HDL-C) levels compared with UK Whites (Ajjan et al., 2007).

Lack of exercise is a risk factor for diabetes Type 2. Physical activity (Gill and Malkova, 2006;Gill and Cooper, 2008), besides cardiorespiratory fitness (Hall et al., 2010;Ghouri et al., 2013), adiposity (Gupta et al., 2010;Shah and Kanaya, 2014) and diet (Lovegrove, 2007;Gupta et al., 2010;Garduno-Diaz and Khokhar, 2012) are life style factors that may influence insulin resistance, glycaemia and diabetes risk.

Physical activity and cardiorespiratory fitness strongly influences glucose tolerance and insulin resistance (Gill, 2007;Leite et al., 2009) and is a strong predictor of diabetes risk and all- cause and CVD mortality risks in both men and women (Lee et al., 2010). Cardiorespiratory fitness (VO_{2max}), for instance, has been shown to predict serum levels of insulin and glucose uptake and strongly positively correlate with insulin sensitivity and secretion in lean and healthy individuals (Larsen et al., 2012a). Physical activity/exercise training significantly improved insulin sensitivity (by 23%) in daughters of patients with type 2 diabetes but not significantly (only 7% increase in insulin sensitivity index) in women with no family history of the disease (Barwell et al., 2008).

The relationship between insulin sensitivity and physical activity is not as strong as the relationship between insulin sensitivity and cardiorespiratory fitness. Lower cardiorespiratory fitness (VO_{2max})levels for instance, contributed to more than two-thirds of the ethnic difference in HOMA_{IR} between SA and EU while physical activity contributed less to this difference (Ghouri et al., 2013).

Furthermore, participating in 3 h/wk of vigorous-intensity activity was shown to be associated with a 22% lower risk of Myocardial Infarction among men which was partially explained by the beneficial effects of physical activity on HDL-C, vitamin D, apolipoprotein B, and hemoglobin A1c (Chomistek et al., 2011).

In the UK, South Asians have been shown to be less physically active than Europeans (Yates et al., 2010; Williams et al., 2011a; Williams et al., 2011b;Ghouri et al., 2013). Other studies that investigated and compared leisure time physical activity among South Asians living in the UK and in South Asia and other regions have also observed lower levels of physical activity in South Asians compared with other ethnic groups (Fischbacher et al., 2004; Joshi et al., 2007; Ye et al., 2009). Physical activity was found to be associated with acute myocardial infarction (Joshi et al., 2007) and low levels of physical activity, especially moderate-to-vigorous physical activity explained for higher insulin resistance among South Asians compared to Europeans in the UK (Ghouri et al., 2013). These findings support the possibility that physical inactivity is likely to contribute to higher risk of coronary heart disease in this ethnic group. Because of the known influence of physical activity on insulin resistance (Gill and Malkova, 2006; Gill and Cooper, 2008) as mentioned above, physical activity is therefore a potential confounder in explaining the habitual diet in relation to insulin resistance (HOMA_{IR}).

5.2 Study Objectives and hypothesis

Objectives

The main objective of the study was to determine whether there were differences in the physical activity of South Asian and European males as well as South Asian and European females. To achieve the main objective, the specific objectives were as follows:

- to determine the physical activity level (PAL) of subjects from selfreported physical activity records as a ratio of total energy expenditure (TEE) to basal metabolic rate (BMR)
- to compare the mean or median MET-min/day for the following five main activity categories: sleeping and resting, activities at work, way of going to work (by walking, cycling, driving or by using public transport), leisure and home activities as well as sports activities
- 3. to determine whether physical activity level and METs for sports activities relate to $HOMA_{IR}$ in South Asians and Europeans, respectively
- 4. to determine the relationship between energy expenditure and ${\rm HOMA}_{\rm IR}$ in Europeans and South Asians

Hypothesis

The physical activity level (PAL) of South Asians would be lower than that of Europeans.

5.3 Subjects and Methods

5.3.1 Subjects

The study subjects in this chapter were participants of the major study on dietary glycaemic index, glycaemic load, other carbohydrate-related dietary variables and HOMA_{IR} in South Asians in Glasgow, UK. Study subjects comprised of a total of 111 subjects of which 52 were South Asian (30 males and 22 females) and 59 were Europeans (30 males and 29 females).

The initial aim was to recruit a target sample of a total of 152 subjects of which 76 would be of South Asian ethnicity (38 males and 38 females) and 76 would be Europeans (38 males and 38 females). This was estimated to give 80% power to detect 5% statistical significance in the percentage of energy intake derived from dietary carbohydrate between ethnic groups for each respective gender with an effect size of 0.66 derived from Sevak et al. (1994). Sample size calculations were estimated using G*power 3.1 program (Faul et al., 2007). A total of 142 subjects were recruited for this study. The number of subjects who completed the whole study comprised of a total of 111 subjects of which 52 were South Asian (30 males and 22 females) and 59 were Europeans (30 males and 29 females). Study compliance is described under subtopic 5.4.8.

Recruitment and general criteria of study subjects are detailed in Chapter 2, General Methods. Ethical approval to conduct the major study was granted by the Medical Faculty Ethics Committee, University of Glasgow. Study subjects were healthy volunteers recruited from the local community in Glasgow, UK. All subjects had resided in Glasgow, UK for at least six months and of either South Asian (from Pakistan) or European ethnicity.

5.3.2 Anthropometric measurements

Anthropometric measurements such as height, weight, waist circumference and body fat percent were taken in the morning on subjects wearing light clothing and no shoes using standard methods of measurement as described in Chapter 2, General Methods, Subtopic 2.4 Anthropometric measurements.

5.3.3 Physical activity record

Physical activity records of study subjects were required to achieve the objectives of this study (see 5.2 Study objectives). For this purpose, subjects were asked to complete a structured physical activity record (Koebnick et al., 2003) concurrently with their weighed food intake diary for up to seven consecutive days. A minimum of three days of recording was deemed acceptable provided that a weighed food record was also completed for three days.

The physical activity record (Appendix VII) consisted of activities grouped into four main categories as follows: sleeping time and rest periods, activities at work (time spent sitting, standing, walking and description of way to work), leisure time and home activities as well as sports. Subjects were asked to mark the activity they performed in the activity record and record how long they spent doing the activity. If activities were not listed in the record; subjects were instructed to write the activity in the box "not listed activities". When carrying out sports activities, subjects were asked to only report the time they were active and record breaks as "leisure activity". Subjects were highly encouraged to fill in the physical activity record immediately or shortly after the activity to avoid forgetfulness or misreporting.

5.3.4 Calculation of physical activity level

Physical activity data was entered into a template of the physical activity record as shown in Table 5.1 onto Excel spreadsheet (MicrosoftWord 2007). A Metabolic equivalent task (MET) value from the Compendium of Physical Activities (Ainsworth et al., 2000; Ainsworth et al., 2011) was assigned to each activity in the physical activity record (Table 5.1). For each subject, the number of hours spent doing each activity was multiplied by the MET value for that activity to obtain a weighted factor for that particular activity. The weighted factor for every activity performed during the day was summed up and then divided by 24 hours to obtain the physical activity level (PAL) for that day. The PAL was averaged over the number of days physical activity was recorded.

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5.3.5 Calculation and determination of occupational and non-occupational activity level and category

Subjects were categorised into occupational activity level categories of either light, moderate, or moderate/heavy by gender (Department of Health., 1991) based on their weighted physical activity factor expressed as METs for the activity multiplied by the time spent (hours) performing the activity. The subjects were also categorised into categories of non-occupational activity (inactive, moderately active or very active) based on their average calculated physical activity level (PAL) as an expression of TEE/BMR (Department of Health., 1991).

5.3.6 Calculation of average total daily METS-min and Energy Expenditure for activities

Daily METS-min/day for a main activity

Each activity in the physical activity fell within one of the five main activity categories such as sleeping and resting, activities at work, way of going to work, leisure and home activities as well as sports activities. For each subject, the MET for each activity was multiplied by the duration (in minutes) the activity was performed and these were averaged over the number of days physical activity was recorded. To obtain the daily METS-min/day for a main activity, the averages of all activities under a particular main activity was summed up.

Accumulated METS-min/week for sports activities

To obtain the accumulated METS-min for sports in a week, the product of the MET score for the particular sports activity multiplied by time spent performing it (in minutes) was summed up for all sports activities that were performed in the week. Sports activities in METS-min/week only included data from subjects who provided 6 to 7 days of physical activity.

Daily total energy expenditure

To obtain the daily energy expenditure of each main activity like sleeping and resting, activities at work, way of going to work, leisure and home activities as well as sports activities, respectively, the MET score for each activity under a main activity was multiplied by the time spent (minutes) on each activity. The

product of this was summed up for all activities under each main activity and multiplied by the basal metabolic rate (BMR) of each individual which was predicted using Oxford-equations (Henry, 2005) and divided by 24hrs (in minutes). The total daily energy expenditure was the sum of energy expenditure of all main activities.

5.3.7 Statistical analysis

Statistical analyses were performed with IBM SPSS Statistics 21 (IBM Corp., Armonk, NY). Descriptive statistics of the study participants' characteristics were computed and presented. All variables for which statistical analyses were carried out on were examined for normality (Shapiro Wilks statistics test for normality) and outliers (Box-plot or box-and-whisker plots). The means for variables between the groups were compared using Independent Samples T-test (P<0.05) while the medians of the variables that were not normally distributed were tested with the Independent Man-Whitney U test. Tables were presented separately by gender.

5.3.7.1 Correlation analysis

The relationship of physical activity variables to HOMA_{IR} was tested with Spearman correlation two-tailed test and Spearman Partial correlation test as these were the most appropriate non-parametric correlation tests (Sheskin, 2004) for non-normally distributed data as with the case of most of the tested variables in the present study. Spearman Partial correlation tests were conducted by SPSS syntax script command (IBM, 2014) in IBM SPSS Statistics 21 (IBM Corp., Armonk, NY). Spearman partial correlation tests statistically controlled for variables that correlated significantly with HOMA_{IR} or known confounders (energy intake, body mass index, age, Scottish Index of Multiple Deprivation). Correlation test results with and without statistically controlling for certain variables are presented in the results section.

Table 5.1: Metabolic Equivalent (MET) values assigned to each activity in the
physical activity record used in the current study

	Activities			METs
	*Sleeping time a	and rest period	S	1.00
-				
Activities	Sitting	light work	(e.g. desk-work, activities on the computer)	1.50
at work		moderate v	work (e.g. forklift truck driving, cashier)	2 50
	Standing	light work	(e.g. salesperson in a store, working in a Lab)	2.50
		Moderate v	vork (e.g. filling shelves in a store)	3 30
		Heavy work	(e.g. masonry work)	4.00
	Walking	Slowly		2.00
		Briskly		2.00
		Carrying so	mething (e.g. a tray, dishes)	3.50
		Carrying he	eavy items (e.g. boxes, containers)	4.00
	Way to work	Walking	Slowly	5.00
	-		Briskly	<u> </u>
		Bicycling	Slowly ¹ (<15 km/h)	4.00
			Moderate ² (15-20 km/h)	8.00
			Fast ³ (20-23 km/h)	10.00
			Very fast ⁴ (23-26km/h)	12.00
		Driving car		2.50
		Travelling	in a bus, train or car	1.30
Not listed				-
	C. I		T. /	
Leisure time	Sedentary activi	ities (e.g. eatir	ig, reading, I v viewing; phoning, computer games)	1.30
& home	Standing activit	ies (e.g. self ha	air dressing), walking slowly (<4 km/h), shopping	2.10
Activities	Light home activ	vities (e.g. coo	king, ironing, dusting), playing a musical instrument	2.30
	Food shopping,	child care, dog	walking, fast walking, (4-6 km/h)	2.80
	Moderate home	activities and	gardening (e.g. hovering, lawn cutting)	4.00
	Home decoratin	g and repairs (e.g. painting, tiling, wall papering)	4.50
	Heavy home act	ivities and gare	dening (e.g. digging, shovelling snow)	5.50
	Very heavy hom	e activities and	gardening (wood-chopping, carrying heavy logs)	6.50
Not listed				
Sports	Bowling, playing	g billiards or po	ol, throwing darts	2 80
Sports	Light gymnastic	s, bicycling-slo	wly, horseback riding, table tennis, volleyball	4 00
	Golfing, dancing	į		4.00
	Aerobics, baske	tball, bicycling	-moderate, rambling	4.JU
	Badminton, indo	oor skating, rov	ving, skiing, tennis	7.00
	Hill climbing, fo	otball, handba	ll, jogging, bicycling-fast, swimming	7.00
	Judo, bicycling-	racing, squash		9.00
	Sports not listed	1:		11.00

5.4 Results

5.4.1 Characteristics of males

Male subjects were approximately the same age (median of 29 and 26 years, respectively) and of similar socioeconomic status as shown by their Scottish Index of Multiple Deprivation rank (Table 5.2). The ratio of employed to unemployed males, respectively, was approximately equal at 50:50 for both ethnic groups. Most South Asian males were married (53%) whereas most European males were single (10%). A majority of male subjects in both ethnic groups were non-smokers. With regard to physical characteristics, body mass index (normal weight category) and waist circumference of male subjects were similar but South Asian males, on average, possessed significantly more body fat (18.95% body fat) than their counterparts (15.28% body fat).

The light occupational activity category comprised of subjects who worked in professional, technical, administrative/managerial and clerical jobs. It also included sales representatives and those who were unemployed. The moderate occupational activity category included students, sales workers and service workers (Table 5.3 and Table 5.4).

A majority of the male subjects had occupations considered to be moderate activity occupations (60% of total male subjects) while the rest had light activity occupations (Table 5.3). No subjects had jobs that were considered to be moderate-heavy activity jobs. A majority of South Asian males had occupations which fell under the moderate activity category (73.3% of South Asian males) while the remaining 26.7% fell under light occupational activity category. In contrast to this, almost equal numbers of European males had occupations considered to be in the light and moderate activity category respectively (53.3% and 46.7% of European males). South Asian and European males in light activity occupations were considered to be very active in their non-occupational activities based on their physical activity levels of 1.58 and 1.61 respectively. All males (South Asians and Europeans) in moderate activity occupations were

considered to be moderately active in their non-occupational activities with similar physical activity level (1.7).

Most female subjects worked in occupations assumed to have moderate occupational activity level. A majority of European females (72% of European females) worked in moderate activity level occupations. South Asian females were categorised as moderately active in their non-occupational activity level while European females in contrast, were categorised as very active.

5.4.2 Physical activity of males

South Asian and European males were both physically active, similar in their physical activity level (PAL) which had a median of 1.61 and 1.60, respectively. European males exercised almost twice as frequently as South Asian males based on their answers to exercise frequency and duration in a separate questionnaire from the physical activity record. In that questionnaire, European males also reported that they exercised for an hour on average and this was approximately 22 minutes longer than the duration reported by South Asian males (Table 5.5).

Table 5.5 shows the mean or median METS-min/day for five main activity categories. The METs-min/day for of all activities in total was similar among males (P-value of 0.554) but their METS-min/day for occupation, leisure time and home as well as sports activities differed significantly (P-value of 0.000, 0.000 and 0.004, respectively). On average, South Asian males expended significantly more MET-min/day at their occupation than European males (947.43 MET-min/day and 449.71, respectively).

However, South Asian males tended to be 20% less physically active than European males in non-occupational activities barring sleeping time and rest periods. This was because compared to European males, South Asian males expended less energy and time in leisure/home activities (360 MET-min/day). South Asian males were also considerably less physically active in sports (median (IQR) of 0.00 (171.00) MET-min/day). South Asian males accumulated significantly less METS-min for sports activities over the week, by 46% less than European males (Table 5.5).

5.4.3 Characteristics of females

South Asian and European females were comparable in their age (median of 27 and 24 years old respectively) and socio-economic status (Table 5.4). Most females were non-smokers (95.5% and 86.2%, respectively) and a majority of them were unemployed (including students). By ethnicity, a higher percentage of employed females were of European ethnicity. Half of South Asian females were married while a majority of European females were single (93%).

With regard to physical characteristics, their median BMI were similar (P-value= 0.106) but South Asian females, compared to European females, tended to have higher BMI (median of 25.25 Kg/m² and 23.40 Kg/m² respectively). Average body fat percent and waist circumference of South Asian females were not significantly different from European females (body fat of 30.65 % and 27.55%, respectively and waist circumference of 78.74cm and 76.26cm, respectively).

5.4.4 Physical activity of females

As shown in Table 5.7, South Asian females were observed to have lower mean physical activity level (1.57) than European females (1.66) and this almost reached statistical significance (0.052). On average, South Asian females exercised a fewer number of times and at shorter durations each time (median of 0) than European females (median of 3 times per week; 45 minutes). Compared to European females, the mean MET-min/day for total activities expended throughout the week by South Asian females was 5.8% lower and this difference almost reached statistical significance (*p*-value=0.052).

South Asian females were found to be considerably less active in sports compared to European females (South Asian females were 57% less active based on mean figures of METS-min/day of sports activities). South Asian females also accumulated significantly less METS-min for sports activities over the week, by 83% less than European females (Table 5.7). Overall, females were similar in MET-min/day expenditure for all other activities such as sleeping and resting, work/occupation, getting to and from work and leisure time and home activities. Out the five activity categories, females of both ethnicities expended the highest MET-min for leisure time and home activities which contributed approximately 46% to 48% of the mean MET-min/day for all activities in total while they expended the least MET-min/day for sports activities.

5.4.5 South Asians and Europeans

South Asians and European males and females combined, respectively, did not differ in demographic characteristics, age and physical characteristics but South Asians had significantly 6% higher fasting insulin than Europeans, were more insulin resistant and had higher CRP levels. South Asians also exercised less frequently, in less duration and expended less energy in sports activities (Table 5.8).

In the group with no reported family history of diabetes (Table 5.9), South Asians differed only in age (South Asians were 6 years older on average), frequency of exercise, exercise duration and daily energy expended for sports for which was lower in South Asians than in Europeans. All other physical and metabolic characteristics did not differ significantly between South Asians and Europeans. In the subset of South Asians and Europeans with reported family history of diabetes, the only variables that differed between the ethnic groups were frequency of exercise, exercise duration and daily energy expended for sports for which was lower in South Asians than in Europeans.

5.4.6 Correlation of physical activity level, sports energy expenditure and daily energy expenditure with HOMA_{IR}, fasting Insulin and fasting glucose in South Asians and Europeans

In both South Asians and Europeans, daily energy expenditure for total activities was found to inversely relate to fasting insulin (Figure 5.1). In South Asians, total energy expenditure related inversely with HOMA_{IR} and this almost reached statistical significance (r= -0.262; p=0.060) but was not related to fasting glucose (Table 5.10). In Europeans, there was a moderate inverse relationship

between total energy expenditure and HOMA_{IR} (r= -0.337; p=0.009) and fasting glucose.

5.4.7 Correlation of physical activity level, sports energy expenditure and daily energy expenditure with HOMA_{IR}, fasting Insulin and fasting glucose in South Asians and Europeans stratified by reported family history of diabetes

South Asians who reported family history of diabetes, but not in those with no family history of diabetes, were observed to have daily energy expenditure for total activities that inversely moderately related to HOMA_{IR} (r= -0.367; p= 0.033) (Table 5.11 and Figure 5.3) and fasting insulin respectively (r= -0.376; p= 0.028) (Table 5.11). Energy expended for sports and physical activity level did not relate significantly to HOMA_{IR} nor fasting insulin. The PAL, sports energy expenditure and total energy expenditure of Europeans with and without reported family history of diabetes were not shown to relate significantly with HOMA_{IR} and Insulin, respectively (Table 5.11, Figure 5.2 and 5.3).

5.4.8 Study compliance

The number of days physical activity records were collected and the percentage of subjects who completed those records in the given days are as follows: South Asian males (n=30), 5-7 days, 77%; 3-4 days, 23% European males (n=30), 5-7 days, 87%; 3-4 days, 13% South Asian females (n=22), 5-7 days, 68%; 3-4 days, 32% European females (n=29), 5-7 days, 76%; 3-4 days, 24%

Variable	I	२)	[†] <i>p</i> -value		
	South Asian males Eur (<i>n</i> 30)		Europea (<i>n</i> 3	n males 30)	
Age (years)	29.00	(7.00)	26.00	(10.50)	(0.276)
SIMD	3729.97	1288.56	3297.77,	1764.93	0.283
Body Mass Index (kg/m ²)	23.75	(3.70)	23.05	(3.55)	(0.290)
Body Fat (%)	18.95,	6.33	15.28,	3.94	0.009
Waist circumference (cm)	83.20 (14.91)		80.65	(7.55)	(0.446)
		Preval	ence (%)		
Employed	50.0		53.3		
Married	53.3		10.0		
¹ NonSmokers	80	6.7	83	.3	

Table 5.2: Characteristics of South Asian and European males

SD, standard deviation; IQR, interquartile range; SIMD, Scottish Index of Multiple Deprivation rank

[†]P-value not in brackets was calculated by Independent Samples T-Test for normally distributed variables; P-value in brackets was calculated by Independent Man-Whitney U test for non-normally distributed variables;

Statistical significance was set at p<0.05

¹ one South Asian subject did not report his status

Table 5.3: Classification of South Asian and European males based on occupational work and non-occupational activity

*Occupational activity category	Total n of sub wit ethnic	umber ojects hin ity (%)	Total (<i>n</i> 60) (%)	Non-occupational activity lev (PAL)	
	SA (<i>n</i> 30)	EU (<i>n</i> 30)		SA (n 30)	EU (n 30)
Light	26.7	53.3	40.0	Very active (1.58)	Very active (1.61)
Moderate	73.3	46.7	60.0	moderately active (1.70)	moderately active (1.70)
Moderate- Heavy					

SA, South Asian; EU, European; *n*, sample size; PAL, Physical activity level

*occupation by activity level:

Light: professional and technical workers; administrative and managerial; Sales Representatives, Clerical and related workers; Unemployed

Moderate: Students; Sales workers; Service workers

Table 5.4: Classification of South Asian and European females based on occupational work and non-occupational activity

*Occupational activity level	Total number of subjects within ethnicity (%)		Total (<i>n</i> 51) (%)	Non-occupatio (P	nal activity level AL)
	SA (<i>n</i> 22)	EU (<i>n</i> 29)		SA (n 22)	EU (n 29)
Light	45.5	27.6	43.1	Moderately active (1.53)	Very active (1.61)
Moderate	54.5	72.4	56.9	Moderately active (1.59)	Very active (1.68)
Moderate- Heavy					

SA, South Asian; EU, European; *n*, sample size; PAL, Physical activity level;

*occupation by activity level:

Light: professional and technical workers; administrative and managerial; Sales Representatives, Clerical and related workers; Unemployed; Housewives

Moderate: Students; Sales workers; Service workers

Table 5.5: Mean and median daily METS-min for five main activity categories and total daily energy expenditure of South Asian and European males

	Ν	[†] <i>p</i> -value			
Main activity categories	South Asian n	nales (<i>n</i> 30)	European ma		
Sleeping time and rest periods (MET-min/day)	486.75	(71.25)	516.43	(94.28)	(0.096)
Work/occupation (MET-min/day)	947.43,	302.75	449.71,	332.59	0.000
Way to work (MET-min/day)	154.09	(167.38)	91.61	(162.78)	(0.147)
Leisure time and home (MET-min/day)	624.92,	299.00	984.95,	316.28	0.000
Sports (MET-min/day)	0.00	(171.00)	126.43	(347.63)	(0.004)
¹ Sports (MET-min/week)	882.56	(481.38)	1626.66	(342.26)	(0.004)
Total activities (MET-min/day)	2321.95	(484.40)	2295.93	(523.84)	(0.554)
Energy Expenditure for total activities (kJ Kg ⁻¹ body mass)	162	(31)	160	(33)	(0.492)
² Physical Activity level (PAL)	1.61	(0.34)	1.60	(0.36)	(0.879)
³ Number of times subjects exercise per week	1.25	(3.00)	3.00	(3.00)	(0.002)
³ Exercise duration each time (min)	25.00	(67.50)	60.00	(61.25)	(0.027)

MET, Metabolic equivalent task; SD, standard deviation; IQR, interquartile range

¹values shown are mean and standard error of mean

²calculated from subjects' self-reported physical activity records

³reported by subjects in a questionnaire separate from physical activity records

[†]*p*-value not in brackets was calculated by Independent Samples T-Test for normally distributed variables; *p*-value in brackets was calculated by Independent Man-Whitney U test for non-normally distributed variables; Statistical significance was set at p < 0.05

Variable		[†] P-value			
	Sout fen (r	h Asian nales n 22)	Euroj fema (<i>n</i> 2	oean ales 29)	
Age (years)	26.50	(11.50)	24.00	(11.50)	(0.439)
SIMD	4308.0 0	(2004.25)	4448.00	(2739.50)	(0.790)
Body Mass Index (kg/m ²)	25.25	(9.18)	23.40	(6.15)	(0.106)
Body Fat (%)	30.65,	9.42	27.55,	8.69	0.230
Waist circumference (cm)	78.74, 12.16		76.26,	10.04	0.429
		l			
Employed	18.2		37	.9	
Married	5	0.0	6.	9	
¹ NonSmokers	ç	5.5	86	.2	

Table 5.6: Characteristics and demographics of South Asian and European females

SD, standard deviation; IQR, interquartile range; SIMD, Scottish Index of Multiple Deprivation rank

[†]*p*-value not in brackets was calculated by Independent Samples T-Test for normally distributed variables; *p*-value in brackets was calculated by Independent Man-Whitney U test for non-normally distributed variables

Statistical significance was set at p < 0.05

¹ one South Asian subject did not report his status

Table 5.7: Mean and median daily METS-min for five main activity categories and total daily energy expe	nditure
of South Asian and European females	

	n	' <i>p</i> -value			
Main activity categories	South Asiar	n females	European	females	
	(<i>n</i> 2	2)	(<i>n</i> 2	9)	
Sleeping time and rest periods (MET-min/day)	553.29,	92.40	533.00,	68.96	0.373
Work/occupation (MET-min/day)	473.57	(564.32)	442.50	(463.81)	(0.711)
Way to work (MET-min/day)	102.98	(216.44)	102.86	(192.97)	(0.886)
Leisure time and home (MET-min/day)	1083.77,	366.63	1107.69,	397.98	0.827
Sports (MET-min/day)	0.00	(3.00)	67.50	(188.40)	(0.012)
¹ Sports (MET-min/week)	225.81	201.27	1313.95	414.36	0.024
Total activities (MET-min/day)	2253.42,	166.23	2393.55,	325.14	0.052
Energy Expenditure for total activities (kJ Kg ⁻¹ body mass)	144.09	20.31	151.07	25.21	0.293
² Physical Activity level (PAL)	1.57,	0.12	1.66,	0.23	0.052
³ Number of times subjects exercise per week	0.00	(1.00)	3.00	(3.25)	(0.000)
³ Exercise duration each time (min)	0.00	(22.50)	45.00	(30.00)	(0.000)

MET, Metabolic equivalent task; SD, standard deviation; IQR, interquartile range

¹values shown are mean and standard error of mean

²calculated from subjects' self-reported physical activity records

³reported by subjects in a questionnaire separate from physical activity records

[†]*p*-value not in brackets was calculated by Independent Samples T-Test for normally distributed variables; *p*-value in brackets was calculated by Independent Man-Whitney U test for non-normally distributed variables;

Statistical significance was set at p<0.05

Table 5.8: Physical, metabolic characteristics and daily energy expenditure of all study subjects (n=111)

Variable		Median	(IQR)		
	South Asia	ans (<i>n</i> 52)	Europea	ans (<i>n</i> 59)	[†] <i>p</i> -value
Age (years)	28.5	(10.3)	24.0	(10.0)	0.168
Scottish Index of Multiple Deprivation	3997.0	(1669)	3978.0	(2825)	0.484
Body Mass Index (kg/m ²)	24.2	(5.35)	23.1	(4.1)	0.073
Body Fat (%)	21.5	(13.9)	18.8	(12.7)	0.090
Waist circumference (cm)	81.3	(12.9)	79.2	(11.9)	0.252
Energy intake (kJ)	8241.4	(2111.7)	8169.0	(3838.8)	0.901
Fasting glucose (mmol/L)	4.84	(0.52)	4.73	(0.72)	0.159
Fasting insulin (μ U/L)	4.94	(3.66)	4.66	(2.49)	0.023
HOMA _{IR}	1.10	(0.93)	0.97	(0.55)	0.005
CRP (mg/L)	0.65	(1.72)	0.34	(0.57)	0.008
¹ Physical Activity level (PAL)	1.6	(0.2)	1.6	(0.33)	0.677
² Number of times exercise /week	0	(2)	3	(3)	0.000
² Exercise duration each time (min)	0	(44)	53	(30)	0.000
Daily energy expended for sports (kJ kg ⁻¹ body mass)	0	(3)	7	(21)	0.000
Total daily energy expenditure (kJ kg- ¹ body mass)	71	(30)	88	(41)	0.831

IQR, Interquartile range

[†]*p*-value for Mann-Whitney U test (for non normally distributed data)

Statistical significance was set at p < 0.05

¹calculated from subjects' self-reported physical activity records

²reported by subjects in a questionnaire separate from physical activity records

Variable	No reported family history of diabetes					Family history of diabetes				
	SA (n 18)		EU (n 44)		P-value	EU (n 34)		SA (n 15)		<i>p</i> -value
Age (years)	30.0	(10.8)	24.0	(8.0)	(0.017)	27.5	(8.3)	27.0	(21.0)	(0.447)
SIMD	4308.0	(1455.3)	3889.0	(2744.3)	(0.675)	4006.9	1313.8	3556.1	2085.9	0.449
Body Mass Index (kg/m ²)	23.7	(3.5)	23.2	(4.0)	(0.443)	25.1	4.7	23.6	4.1	0.309
Body Fat (%)	19.8	(7.2)	17.1	(11.3)	(0.261)	25.0	10.2	23.5	9.4	0.647
Waist circumference (cm)	84.0	(21.3)	79.2	(10.8)	(0.059)	80.7	11.2	79.5	9.5	0.720
Energy intake (kJ)	8909.3	(1810.5)	8069.7	(4095.6)	(0.411)	7991.1	(2278.9)	8171.0	(3507.1)	(0.729)
Fasting glucose (mmol/L)	4.81	0.45	4.70	0.42	0.325	4.91	0.41	4.92	0.55	0.957
Fasting insulin (μU/L)	4.81	(2.25)	4.70	(2.44)	(0.258)	5.0	(5.4)	4.3	(4.8)	(0.129)
HOMA _{IR}	1.07	(0.55)	0.98	(0.55)	(0.119)	1.11	(1.01)	0.93	(1.30)	(0.129
CRP (mg/L)	0.40	(1.46)	0.32	(0.59)	(0.306)	1.08	(2.07)	0.39	(0.85)	(0.097)
¹ Physical Activity level (PAL)	1.6	(0.4)	1.6	(0.3)	(0.895)	1.6	(0.2)	1.7	(0.4)	(0.558)
² Number of times exercise per week	0.0	(3.5)	3.0	(2.9)	(0.033)	0.0	(2.0)	3.5	(3.5)	(0.000)
² Exercise duration each time (min)	0.0	(30.0)	45.0	(38.8)	(0.002)	0.0	(60.0)	60.0	(45)	(0.004)
Daily energy expended for sports (kJ kg ⁻¹ body mass)	0	(0)	6	(14)	(0.006)	0.0	(7.5)	9.0	(43)	(0.004)
Total daily energy expenditure (kJ kg ⁻¹ body mass)	155	(31)	150	(34)	(0.614)	153	23	152	25	0.869

Table 5.9: Physical, metabolic characteristics and daily energy expenditure of all study subjects (n=111) stratified by family history of diabetes

SD, standard deviation; IQR, interquartile range; ¹calculated from subjects' self-reported physical activity records; ²reported by subjects in a questionnaire separate from physical activity records; [†]*p*-value not in brackets was calculated by Independent Samples T-Test for normally distributed variables; *p*-value in brackets was calculated by Independent Man-Whitney U test for non-normally distributed variables; Statistical significance was set at *p*<0.05

Table 5.10: Correlation of physical activity level, sports energy expenditure and daily energy expenditure with $HOMA_{IR}$, fasting Insulin and fasting glucose in South Asians and Europeans

	South f	n Asian male emales (n 5	es and 2)	European males and females (n 59)			
Variables	HOMA _{IR}	Insulin <i>r</i> (p-value)	Glucose	HOMA _{IR}	Insulin <i>r</i> (p-value)	Glucose	
Physical Activity level	-0.065	-0.100	0.066	-0.229	-0.172	-0.154	
	(0.648)	(0.482)	(0.644)	(0.081)	(0.191)	(0.246)	
Sports	-0.060	-0.140	0.235	-0.171	-0.161	0.031	
(kJ Kg ⁻¹ body mass)	(0.671)	(0.323)	(0.094)	(0.196)	(0.223)	(0.814)	
Total activities	-0.262	-0.300	0.059	-0.337	-0.268	-0.292	
(kJ Kg ⁻¹ body mass)	(0.060)	(0.031)	(0.676)	(0.009)	(0.040)	(0.025)	

HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance; n, sample size

r Spearman correlation coefficient

Statistically significant correlation was set at p<0.05

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Table 5.11 : Correlation of physical activity level, sports energy expenditure and daily energy expenditure with HOMA_{IR}, fasting Insulin and fasting glucose in South Asians and Europeans stratified by reported family history of diabetes

	No family history of diabetes				Family history of diabetes				
	South Asians (n 18)		Europeans (<i>n</i> 44)		South Asians (<i>n</i> 34)		Europeans (<i>n</i> 15)		
	HOMA _{IR} <i>r</i> (p-value)	Insulin <i>r</i> (p-value)	HOMA _{IR} <i>r</i> (p-value)	Insulin <i>r</i> (p-value)	HOMA _{IR} <i>r</i> (p-value)	Insulin r (p-value)	HOMA _{IR} <i>r</i> (p-value)	Insulin r (p-value)	
Variables					, ,,	, ,,			
Physical Activity level	0.304 (0.220)	0.167 (0.507)	-0.179 (0.244)	-0.125 (0.421)	-0.216 (0.220)	-0.210 (0.232)	-0.262 (0.346)	-0.138 (0.624)	
Sports activities (kJ Kg ⁻¹ body mass)	0.180	-0.002	-0.152	-0.149	-0.181	-0.223	0.147	0.118	
	(0.476)	(0.995)	(0.324)	(0.336)	(0.304)	(0.205)	(0.600)	(0.674)	
Total activities (kJ Kg ⁻¹ body mass)	0.147	-0.001	-0.277	-0.198	-0.367	-0.376	-0.180	-0.080	
	(0.562)	(0.997)	(0.068)	(0.197)	(0.033)	(0.028)	(0.520)	(0.777)	

HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance; *n*, sample size

r Spearman correlation coefficient

Statistically significant correlation was set at p < 0.05



Figure 5.1: Relationship between energy expenditure (kJ-1 body mass) and HOMA_ $_{\rm IR}$ in Europeans and South Asians



Figure 5.2: Relationship between energy expenditure (kJ-1 body mass) and $HOMA_{IR}$ in Europeans and South Asians with **no reported family history of diabetes**



Figure 5.3: Relationship between energy expenditure (kJ-1 body mass) and $HOMA_{IR}$ in Europeans and South Asians with reported family history of diabetes

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5.5 Discussion

South Asians are known to be more insulin resistant (Bakker et al., 2013;Ghouri et al., 2013). Physical activity and cardiorespiratory fitness strongly influences glucose tolerance and insulin resistance (Gill, 2007;Leite et al., 2009) and is a strong predictor of diabetes risk in both men and women (Lee et al., 2010). In the UK, South Asians have been shown to be less physically active than Europeans (Williams et al., 2011a). Therefore this difference in physical activity may explain for the higher insulin resistance among South Asians because physical activity is known to influence insulin resistance (Gill and Malkova, 2006;Gill and Cooper, 2008). In other words, physical activity is a potential confounder in making ethnic comparisons (South Asians and Europeans) regarding habitual diet and insulin resistance (HOMA_{IR}).

One of the study aims was to determine whether there were ethnic differences in the physical activity of the study subjects who were involved in the study on dietary glycaemic index, glycaemic load, other carbohydrate-related factors and insulin resistance in South Asians in Glasgow, UK (chapter 6). This was necessary because physical activity, as mentioned previously, influences insulin sensitivity (Gill and Malkova, 2006). Therefore physical activity of study subjects were described in this chapter and accounted for in chapter 6 where correlations of dietary variables and insulin resistance were carried out.

Physical activity records were used to quantify physical activity of all study subjects and compare physical activity of South Asians to Europeans. Physical activity records are practical, relatively easily administrated, less expensive and can be employed to obtain qualitative information on physical activity (Shephard, 2003;Janz, 2006). Self-reported physical activity has been found to be at least modestly correlated with objective measures obtained using criterion methods (Tudor-Locke and Myers, 2001;Aadahl et al., 2007). The study participants were instructed to report their physical activity in order to achieve several objectives listed below. Physical activity level (PAL) in males and females ranged from moderately active to very active. This is because in this study sample, the ratio of employed to unemployed subjects was almost equal and those who were employed were physically active both at their occupation and outside work. Unemployed subjects (not students) were also physically active in non-occupational activities like leisure, home activities as well as sports activities. Students (considered to be unemployed), were physically active in study activities like working in the lab for instance, and walking/cycling to university. Most students were also active in leisure and sports activities. Although South Asians and Europeans by their respective gender, expended similar MET-min/day for total activities, they differed on the MET-min/day spent on non-occupational activities.

Physical activity of males

South Asian males expended significantly more time and energy at their occupation. METS-min expended at work depended largely on the type or nature of their occupation. A shop-keeper who fills shelves in a store while standing for instance, would be considered as doing moderate work of 3.30 METs in contrast to an office clerk who mostly does light work sitting at his desk or working on the computer which has a METs of 1.50. The higher MET-min/day for activities at work in South Asian males compared to European males was due to their occupation which required them to do more moderate intensity (METs) work as opposed to light intensity work. Although the time spent at work was comparable in both ethnic groups, South Asians spent less time in non-occupational activities such as sports as well as leisure time and home activities.

South Asian males evidently exercised less frequently and at shorter durations than European males based on what they reported in a questionnaire separate from physical activity records. The lower MET-min/day that South Asian males spent in sports activities compared to European males confirmed the South Asians' reported frequency and duration of exercise which were found to be less than those reported by European males. This however, did not translate to a lower physical activity level (PAL) in South Asian males nor in lower METSmin/day for total activities which was in fact, observed to be similar to that of the European males. This is because PAL calculation and METS-min/day for total activities is not only derived from sports and exercise activities but also includes all other activities the subjects performed throughout the day for up to a week. Participation in sports (which will promote cardiorespiratory fitness) was significantly lower in South Asians. Thus, regardless of the same PAL, South Asians may possibly be less fit since only structured exercises promote cardiorespiratory fitness (Johannsen et al., 2013;Giannaki et al., 2015).

Physical activity of females

Females were similar in MET-min/day expenditure for non-occupational activities like sleeping and resting, work/occupation, getting to and from work and leisure time and home activities, females of both ethnicities had the highest MET-min/day for leisure time and home activities which contributed approximately 46% to 48% of the mean MET-min/day for all activities in total while they expended the least MET-min/day for sports activities.

Physical activity is a problem in South Asian females. South Asian females were observed to have lower PAL levels compared to those of European females and they were also found to be considerably less physically active in sports activities. Furthermore they exercised less frequently and spent less time in exercise. Notably, South Asian females were observed to be 57% less active in sports activities.

South Asian females (and males) were physically less active in more vigorous, higher intensity (METs) activities such as sports, exercise and leisure activities like walking briskly. This may have negative impact on their health, including on insulin sensitivity. Participating in 3 h/wkj⁻¹ of vigorous-intensity activity is associated with a 22% lower risk of myocardial infarction among men (Chomistek et al., 2011).

In this study, South Asians were less physically active then Europeans. One of the reasons may be that, for the South Asian females, 50% of them were married and almost all had children. Perhaps the South Asian females had commitments at home and with family so they had less time to exercise. Cultural barriers, such as religious modesty or avoidance of activities which would involve being at close range to male non-family members as well as fear of going out alone may inhibit participation in exercise, sports and leisure activities (Hayes et al., 2002;Sriskantharajah and Kai, 2007). Cultural attitudes and values held by South Asians in general, do not encourage sports and physical activity (Hayes et al., 2002).

Among South Asian females with CHD and diabetes, they were uncertain about what level of physical activity was appropriate and safe for them so they had their own limits for physical activity in relation to their condition, which they were afraid to exceed (Sriskantharajah and Kai, 2007). However, the study subjects of the current study were all apparently healthy the possible reasons above probably applied to them.

Overall, in this current study, South Asians were less physically active especially in sports, exercise and leisure activities. Thus, their cardiorespiratory fitness can be expected to be lower. These findings are in agreement with previous studies (Hayes et al., 2002;Fischbacher et al., 2004;Becker et al., 2006;Joshi et al., 2007;Yates et al., 2010;Williams et al., 2011a) that also described South Asians to have low physical activity. South Asians were found to be substantially less physically active than Europeans (males and females) as observed by their lower total MET-min/day score for non-occupational activities in comparison to Europeans (Williams et al., 2011a) and this was especially evident in South Asian women.

Total MET-min/day for non-occupational activities (except sleeping and resting) in South Asian males was similar to the study by Williams et al. (2011) where males in that study were found to be less physically active than Europeans by 37%. In the current study, South Asian males were 33% less physically active in non-occupational activities compared to their counterparts. For females in our study, South Asian females' MET-min/day on non-occupational activities was 13% lower than in female Europeans while previous investigations had observed bigger discrepancies of approximately 29% lower (Williams et al., 2011a).

Other studies also showed that migrant and urban South Asians tended to be physically less active (Hayes et al., 2002;Fischbacher et al., 2004;Mohanty et al., 2005). Use of the physical activity questionnaire in this study had several
advantages in that it reduced subject burden due to its organized structure which did not require subjects to write anything besides marking on the spaces corresponding to the amount of time they spent in performing a specific activity. It also addressed spontaneous or routine light and moderate activities, such as household chores, leisure time and occupational activities. This possibly captured activities which may be characteristic of a sedentary population which may be the case for some of the study subjects.

Physical activity level and other components of physical activity (ie. sleeping time and rest, activities at work, leisure time and home activities, sports activities as well as total activities) did not significantly relate to HOMA_{IR} in both South Asians and Europeans with and without statistically controlling for total energy intake, BMI, body fat percent, age and socio-economic status (SID).

The use of the physical activity questionnaire to describe PAL and physical activities expressed as MET-min/day, may however, had some limitations. There was a possibility that study subjects under or over-reported their physical activity. This is because under- or overreporting of physical activity may occur in some populations or individuals which may affect interpretation of the findings and result in biased conclusions (Cleland et al., 2011;Panter et al., 2012).

There may potentially be individual as well as ethnic differences in how study subjects interpreted the activities listed in the physical activity record which would have influenced which activity in the physical activity record would match the activity that they performed. This in turn, would influence their METmin/day averages because each specific activity in the physical activity has a MET-value that may be low, medium or high. For example, the pace for walking may be interpreted differently by individuals and their interpretation would ultimately decide which activity they would record whether "walking briskly" or "walking slowly" for instance, where walking briskly has a higher METs. However, no subjects reported that they were unsure about how to record their physical activity.

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Correlations between physical activity level and components of physical activity

It was apparent that the less the total energy expenditure, the higher the fasting insulin in both South Asians and Europeans. Possibly a lower physical activity confers lower cardiorespiratory fitness levels and this relates to a higher fasting insulin and HOMA_{IR}. Both South Asians and Europeans showed a pattern of lower HOMA_{IR} with increase in daily energy expenditure almost reaching statistical significance (possibly to due to smaller sample size) in South Asians (Figure 5.1). Lower level of physical activity have been reported to be independently associated with HOMA_{IR} or increased risk for the development of insulin resistance in populations of mixed ethnicity in the US (Grandinetti et al., 2015).

When subjects were stratified by ethnicity and reported family history of diabetes, only South Asians with reported family history of diabetes showed inverse relationship between daily energy expenditure and HOMA_{IR} (Figure 5.3) while in all other groups, this was not observed. Perhaps an individual of South Asian ethnicity with reported family history of diabetes is more predisposed to the detrimental impact of low physical activity leading to a higher risk of developing insulin resistance. Similarly, individuals predisposed to insulin resistance such as nondiabetic daughters of patients with type II diabetes, HOMA_{IR} as well as other features of the 'metabolic syndrome' (i.e., waist circumference, fasting glucose, fasting insulin, fasting TG) were found to significantly negatively correlate with daily energy expenditure but these correlations were not as pronounced in individuals with no family history of diabetes (Higgins et al., 2005).

In Europeans, a statistically significant relationship between energy expenditure and HOMA_{IR} was not observed regardless of reported or no reported family history of diabetes. It was previously shown that Europeans were more physically active in overall PAL (in EU females compared to SA females) and that Europeans, compared to South Asians, were more physically active in sports or structured exercise. Higher physical activity confers better cardiorespiratory fitness levels and this may relate to higher insulin sensitivity. Europeans with reported history of diabetes probably benefited from the protective effect of relatively higher physical activity and thus, cardiorespiratory fitness on insulin sensitivity.

5.6 Conclusion

Both South Asian and European males were observed to be physically active at similar physical activity levels but South Asian males were less physically active in non-occupational activities such as leisure and home activities as well as sports and exercise. South Asian females were found to be less physically active compared to European females in general and had lower mean PAL values. South Asian females were considerably less physically active <u>in sports and exercise</u> <u>activities</u>. South Asian ethnicity, having low physical activity and having family history of diabetes may predispose an individual to insulin resistance.

Chapter 6 Dietary glycaemic index, glycaemic load and insulin resistance (HOMA_{IR}) of South Asians in Glasgow, UK and ethnic comparisons (South Asian and European)

The previous three chapters provided information on the diet and physical activity of study subjects which were required to support the study findings in this chapter. In this chapter, we focus on the habitual dietary glycaemic index (GI), glycaemic load (GL) and carbohydrate-related variables in relation to insulin resistance (HOMA_{IR}) in South Asians. These were a healthy sample of South Asians (n 52; 30 males and 22 females) residing in Glasgow, UK at the time of the study recruitment. The habitual dietary GI and GL of study subjects, estimated from weighed food records were likely habitual (Chapter 4). The estimation of dietary GI and GL considered the GI of foods as mixed-meals including the GI values of several key staples in the South Asian diet derived from measurements in our own lab (Chapter 3). In this chapter, physical activity level and/or METS-min/day for non-occupational activities (physical activity of study subjects were presented in Chapter 5) were statistically controlled for in correlation tests between dietary GI, GL and HOMA_{IR}.

6.1 Introduction

The role of the glycaemic index (GI) and/or glycaemic load (GL) of foods in relation to health and disease has been a matter of debate (Hare-Bruun et al., 2008). Prospective cohort studies have shown associations between high GI and/or GL, with an increased risk for some (Ma et al., 2012;Sieri et al., 2013) but not all cardiovascular disease types (Fan et al., 2012). Similarly, low dietary GI and GL are associated with a reduced risk of type 2 diabetes in some (Jakobsen et al., 2010;Greenwood et al., 2013) but not all prospective cohorts (Simila et al., 2011). On the other hand, evidence from randomised control trials has shown that low GI and GL diets are effective in reducing cardiovascular risk factors in overweight and obese subjects (McMillan-Price et al., 2006;Gogebakan et al., 2011;Schwingshackl and Hoffmann, 2013) and in subjects with

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type 2 diabetes (Jenkins et al., 2012). Inconsistencies in the observational evidence have been attributed to the limitations of dietary assessment methods.

The possibility that GI and/or GL relate to health and disease namely diabetes and CVD may be of even more relevance to South Asians as this ethnicity is an established major risk factor for the development of diabetes T2 (Garduno-Diaz and Khokhar, 2012). Currently, diabetes Type 2 is estimated to affect approximately 246 million people worldwide with the highest and fastest growing prevalence in South Asians (Misra et al., 2010b;Garduno-Diaz and Khokhar, 2012). To tackle this problem, much research has been focused on trying to understand the reasons for this high prevalence in South Asians and the risk factors involved.

Among suggested hypothesis/mechanisms explaining for insulin resistance in South Asians include genetic predisposition (thrifty phenotype theory) which claims that in certain environmental circumstances (such as 'feast-or-famine days of hunting and gathering cultures' for instance), the predisposition to diabetes probably evolved as an adaptive trait and this later became disadvantageous when life style changes occurred ('continuous feasting' for instance) (Neel, 1962). Adiposity or overweightness and obesity, physical activity and diet (Bakker et al., 2013) are some other suggested hypothesis. Diet of South Asians in relation to insulin resistance has not been as widely investigated as some of these other factors. Dietary patterns may be associated with adverse metabolic changes which influence health. Pakistanis and Indians who live in Britain are 4-6 times more likely to develop diabetes type 2 than individuals of other ethnic groups in the general population (D'Costa et al., 2000) and this may be influenced by changes in lifestyle after migration (Bhatnagar et al., 1995;Anderson et al., 2005;Gilbert and Khokhar, 2008).

Imbalances in the diet of South Asians for particular macronutrients/nutrients have been associated to insulin resistance. These imbalances include high intake of total dietary fat, saturated fats, polyunsaturated fatty acids (PUFA), trans fatty acids and carbohydrates and low intake of monounsaturated fatty acid, omega-3 PUFAs and fiber (Sevak et al., 1994;Lovegrove, 2007;Misra et al., 2009b). Studies on relationship/correlations of dietary GI and GL to insulin resistance in South Asians are very limited and focused on South Asians living in India (Mohan et al., 2009;Radhika et al., 2009). Findings of that study may not apply to South Asians living in the UK due to differences in environment, acculturation and variation in the food supply. South Asians in India, compared to South Asians in the UK, had carbohydrate intakes that were considerably high (approximately 65.6% (Mohan et al., 2009) versus 45% (Anderson and Lean, 2005) of total energy intake) and their fat intakes also differed considerably (approximately 20 to 26% across quintiles of unadjusted dietary GI (Radhika et al., 2009) versus 40% (Anderson and Lean, 2005). Furthermore those studies did not specifically measure insulin resistance (HOMA_{IR}) and did not compare South Asians to a matched group of white Europeans.

Potential ethnic differences in dietary GI, GL and HOMA_{IR} between South Asians and Europeans were also investigated. Many studies have reported that ethnic groups living in the UK who originate from the Indian subcontinent, when compared with Europeans/white Caucasians, have higher levels of plasma triglycerides (TAG), insulin resistance, C-reactive protein, plasminogen activator inhibitor-1 and lipoprotein (a) and lower levels of HDL-cholesterol (Lovegrove, 2007; Merchant et al., 2007) and substantially higher prevalence of diabetes in this ethnic group (McKeigue et al., 1991) compared to Europeans. Observations of higher insulin resistance (HOMA_{IR}) among South Asians than Europeans in the UK have been reported (Ghouri et al., 2013;Goff et al., 2013). In the study by Ghouri et al., (2013), ethnic differences in HOMA_{IR} were explained by total adiposity in South Asians and lower cardiorespiratory fitness levels (VO₂max) but dietary GI and GL was not discussed in the study. Although the study by Goff et al., reported and compared habitual dietary GI and GL values of South Asians to Europeans; correlation analysis of dietary GI and other nutrient intakes were not the focus of the study and the method of assigning GI values to mixed meals were not clarified.

To the best of our knowledge, there are no published studies which specifically investigate dietary GI, GL and $HOMA_{IR}$ of South Asians and Europeans (from the same locality) in the UK within the wider picture of the whole diet. The habitual diet of South Asians could potentially be similar, lower or higher in glycaemic index and glycaemic load compared to Europeans. This prompted observations

of whether similarities or differences in the habitual dietary GI and GL, taken together with other diet characteristics, physical characteristics, socioeconomic status, physical activity and subjects' reported family history of diabetes and/or CHD could explain for the higher HOMA_{IR} among South Asians compared to Europeans. Habitual dietary GI, GL and HOMA_{IR} of South Asians and Europeans (males and females, respectively) that were recruited for this present study were compared.

6.2 Study objectives and hypothesis

Objectives

- To estimate the habitual dietary glycaemic index and glycaemic load of a sample of South Asian males (n 30) and females (n 22) living in Glasgow, UK and explain for this by describing their carbohydrate food intakes.
- 2. To determine whether dietary GI and GL relate to $HOMA_{IR}$ in South Asians through correlation tests
- 3. To determine the differences/similarities in habitual dietary GI, GL, other dietary variables and HOMA $_{\rm IR}$ in South Asian and European females from the current study
- 4. To determine whether total dietary fat intake, saturated fat, monounsaturated fat and polyunsaturated fat intakes relate to $HOMA_{IR}$ in South Asians through correlation tests

Hypothesis

- 1. The habitual dietary GI of South Asians is higher than the habitual dietary GI of Europeans
- 2. There is a positive relationship between dietary GI and/or GL and $\mbox{HOMA}_{\mbox{\tiny IR}}$ in South Asians

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6.3 Subjects and Methods

6.3.1 Subjects

The study subjects in this chapter were participants of the major study on dietary glycaemic index, glycaemic load, other carbohydrate-related dietary variables and HOMA_{IR} in South Asians in Glasgow, UK. The initial aim was to recruit a target sample of a total of 152 subjects of which 76 would be of South Asian ethnicity (38 males and 38 females) and 76 would be Europeans (38 males and 38 females). This was estimated to give 80% power to detect 5% statistical significance in the percentage of energy intake derived from dietary carbohydrate between ethnic groups for each respective gender with an effect size of 0.66 derived from Sevak et al. (1994). Sample size calculations were estimated using G*power 3.1 program (Faul et al., 2007). A total of 142 subjects were recruited for this study. The number of subjects who completed the whole study comprised of a total of 111 subjects of which 52 were South Asian (30 males and 22 females) and 59 were Europeans (30 males and 29 females). Study compliance is described under subtopic 6.4.3.

Recruitment and general criteria of study subjects are detailed in Chapter 2, General Methods. Ethical approval to conduct the major study was granted by the Medical Faculty Ethics Committee, University of Glasgow. Study subjects were healthy volunteers recruited from the local community in Glasgow, UK. All subjects had resided in Glasgow, UK for at least six months and of either South Asian (from Pakistan) or European ethnicity.

6.3.2 Anthropometric measurements

Height, weight, body fat percentage and waist circumference of subjects were measured using standard equipment and methods (refer to Chapter 2, General Methods, subtopic 2.4 for full description of materials and methods). Body mass index (BMI) was calculated using the standard formula: weight (kg)/height (m²).

6.3.3 Food frequency questionnaire

Food Frequency Questionnaire (FFQ) that has been validated in a South Asian population in the UK (Kassam-Khamis et al., 1999) and adapted to suit the purpose of the study was employed because the primary focus of this research is on South Asians. This FFQ was used to check for study subjects' consistency in reporting carbohydrate food consumption by determining whether carbohydrate food intakes estimated from the FFQ related to intakes estimated from weighed food records using correlation tests. Details on the FFQ, its administration and analysis are described in Chapter 4, subtopic 4.3.3, 4.3.4 and 4.3.5.

6.3.4 Weighed food records

Subjects were required to complete a food inventory or weighed food records (Appendix VI). They were given a digital weighing scale to weigh foods and drinks. Data for subjects who recorded at least three days of food and drink intake, one being a weekend day, were deemed acceptable and included in further analysis. Weighed food records were analysed to obtain average nutrient intakes using diet analysis software, WinDiets 2005 (The Robert Gordon University, Aberdeen, UK). Full details on administration of weighed food records and analysis are described in Chapter 4, subtopic 4.3.6 and 4.3.7.

6.3.5 Relative validation of weighed food records

Weighed food records were examined for relative validity using the Goldberg cut-off method (Goldberg et al., 1991) in line with the guidelines for its calculation and use (Black, 2000) as described in detail in Chapter 4, subtopic 4.3.8.

6.3.6 Determination of habitual carbohydrate food intake (types and amounts) from weighed food records

From weighed food records, the quantity (grams) that were reported to be consumed on each recorded day for the following : 1) carbohydrate foods and 2) foods that were mostly eaten in combination with main staples (rice, chapatti, potatoes, pasta) were entered manually in excel spreadsheet and averaged (grams/day) separately by gender and ethnic group (South Asian, European). This is described in detail in Chapter 4, subtopic 4.3.9.

6.3.7 Determination of dietary glycaemic Index and glycaemic load

6.3.7.1 Glycaemic index value assigned to food items

Each food item recorded in a weighed food records was assigned a mean glycaemic index (GI) value of a food that was most similar to it in terms of its name, description, ingredients, brand, formula, place of product manufacture based on the glucose standard from published values of GI for foods in the United Kingdom (Henry et al., 2005a;Henry et al., 2005b;Henry et al., 2006;Henry et al., 2007;Aston et al., 2008;Henry et al., 2008) and from the international tables for glycaemic index values (Atkinson et al., 2008).

Mixed meals in the weighed food records were assigned a GI value of a mixed meal in a published source such as those listed in the international tables for glycaemic index values (Atkinson et al., 2008) on condition that the meal from the published source was similar to the meal reported in the weighed food records in terms of name (description), ingredients and energy contributed by macronutrients. Curried meals were assigned the GI of the meals derived from our own laboratory measurements (Chapter 3) provided they were similar in name (description), ingredients and energy contributed by macronutrients. For mixed meals where there was no GI value to refer to in the published tables or anywhere else, the GI of the mixed meal was predicted based on the formula by Wolever et al. (1994).

The mixed-meal values from the International GI tables and other publications were used rather than predicted from the ingredients in a mixed meal using the formula by Wolever et al. (1994) because addition of certain toppings to foods (Henry et al., 2006) and varying amounts of fat/oil and protein to a meal have been found to be lower than the GI predicted (Dodd et al., 2011) based solely on the main carbohydrates foods/ingredients in the mixed meal.

6.3.7.2 Calculation of glycaemic Index and glycaemic load

The calculation of average dietary GI and GL from weighed intake dairies was based on the equation described by (Wolever et al., 1994). For each subject, per day, dietary GI was calculated by multiplying the GI value of each food item by the digestible carbohydrate content of the food. The resulting values were then summed up and divided by the total amount of digestible carbohydrate intake per day (Equation 2.1). For the weighed food intake method which involved food recording up to 7 days, average dietary GI for each subject was calculated as the mean GI in the number of days food intake was recorded. Dietary GL was calculated for each subject in the same way as the average dietary GI value but by dividing by 100 instead of the total digestible carbohydrate intake (Equation 2.2).

Equation 6.1: GI per day = Σ (CHO content of food in grams X GI of food) (total amount of CHO in grams)

Equation 6.2: GL per day = Σ (CHO content of food in grams X GI of food) 100

6.3.8 Physical activity record

Subjects were asked to complete a structured physical activity record (Koebnick et al., 2003) concurrently with their weighed food records for up to seven consecutive days. Details on the physical activity record, its administration and analysis are described in Chapter 5, subtopic 5.3.3, 5.3.4, and 5.3.5.

6.3.9 Biochemical analysis

6.3.9.1 Blood collection and plasma preparations

Fasting blood samples were collected into a pre-cooled 9.0 ml ethylenediamine tetra-acetic acid (EDTA) VacutainerTM tube (BD Vacutainer Systems, Plymouth, UK). These blood samples were then centrifuged at 3000 RPM for 15 minutes at 4^{0} C. After centrifugation, aliquots of plasma were transferred using a disposable plastic Pastuer pipette into labeled 1.5-ml Eppendorf tubes (Eppendorf AG, Hamburg, Germany). Plasma were stored at -80^oC until analysis.

6.3.9.2 Plasma insulin measurements

Fasting insulin concentrations were determined using the Mercodia Ultrasensitive Insulin ELISA (Mercodia AB, Sylveniusgatan 8A, Uppsala, Sweden) according to the manufacturers' standard protocol. All plasma samples, before analysis, were thawed to room temperature slowly prior to beginning the assay. Mercodia Insulin ELISA works on the principle that it is a solid phase two-site enzyme immunoassay based on the direct sandwich technique in which two monoclonal antibodies are directed against separate antigenic determinants on the insulin molecule. During incubation, insulin in the samples react with peroxidase-conjugated anti-insulin antibodies and anti-insulin antibodies bound to a microtitration well. A washing step removes unbound enzyme labelled antibody. The bound conjugate is detected by reaction 3,3',5,5'tetramethylbenzidine (TMB). The reaction is stopped by adding a stop solution $(0.5 \text{ MH}_2\text{SO}_4)$ to give a colorimetric endpoint. This was then read spectrophotometrically at 450 nm within 30 min using Multiskan spectrum spectrophotometer (Thermo Fisher Scientific, Surrey, UK) and from these OD readings, fasting insulin was determined by the software program in Multiskan

spectrum spectrophotometer. The coefficient of variation (CV%) for fasting plasma insulin readings was 3.81%.

6.3.9.3 Plasma glucose measurement

Fasting plasma glucose were measured using a modified version of that described for the Glucose Liquicolor (GOD-PAP) Method without Deproteinisation (Human Gesellschaft fur Biochemica und Diagnostica mbH, Germany). The test principle is that the glucose in the samples is determined after enzymatic oxidation in the presence of glucose oxidase (in the reagent). The hydrogen peroxide that is formed reacts under catalysis of peroxidise with phenol and 4-aminophenazone to a red-violet quinoneimine dye as indicator. The following is a description of the method for glucose measurement. All plasma samples to be analysed were thawed to room temperature slowly prior to beginning the assay. Sernorm Quality Control was prepared by pipetting 5ml of distilled water onto the Seronorm powder (this is dissolved within 30 minutes by occasional gentle swirling, avoiding formation of foam. Standards were prepared by making serial dilutions of the 5.55mmol/L standard glucose solution which is provided.

The standards were prepared by pipetting as follows: Standard 4: 1 ml of 5.55mmol/L stock standard provided Standard 3: 500µl of standard 4 + 500 µl distilled water Standard 2: 500µl of standard 3 + 500 µl distilled water Standard 1: 500µl of standard 2 + 500 µl distilled water

Each of the serial dilutions of the standard were mixed using a vortex mixer for 30 seconds. Using the 96-well plate plan, 10 μ l of the glucose reagent were pipetted into wells A1 and A2, 10 μ l of standards 1 to 4 were pipetted into wells B1/B2 to E1/E2, 10 μ l of Sernorm quality control were pippeted into wells F1/F2 and 10 μ l of samples were pipetted into subsequent wells according to the plan. Then 200 μ l of glucose liquicolor reagent were pippetted into each well using a multichannel pipetter. The plate was brought to the spectrophotometer and mixed for 10 seconds on high for 3 times. Then the plate was incubated for 5 min at 37°C in the spectrophotometer. The absorbance of the standards and samples were then measured against the reagent blank at optical density

500nm. The glucose value for the quality control was ensured to be within the acceptable range of 3.81 to 5.27 mmol/L. The coefficient of variation (CV%) for fasting plasma glucose readings was 2%.

6.3.9.4 Plasma Human C-Reactive Protein Immunoassay measurements

Human C-Reactive Protein (CRP) were determined using the Quantikine Human CRP Immunoassay. In principle, this assay uses the quantitative sandwich enzyme immunoassay technique. A monoclonal antibody specific for CRP has been pre-coated onto a microplate. Standards and samples are pipetted into the wells according to the manufacturers' standard protocol and any CRP present is bound by the immobilized antibody. After washing away any unbound materials, an enzyme-linked monoclonal antibody specific for CRP is added to the wells. After a wash to remove any unbound antibody-enzyme reagent, a substrate solution was added to the wells and colour develops in proportion to the amount of CRP bound in the initial step. The colour development is stopped and the intensity of the colour is measured.

All plasma samples to be analysed were thawed to room temperature slowly prior to beginning the assay. The reagents, samples and standards were prepared as instructed by the assay manufacturers. 100µL Assay Diluent RD1F was added to each well then 50 µL of standard, control or sample was added to each well and incubated for 2 hours at room temperature. After incubation, samples were aspirated and washed 4 times and 200 µL of Conjugate was added to each well and incubated for 2 hrs at room temperature. Following this, samples were aspirated and washed 4 times and 200 µL of Substrate Solution was added to each well and incubated for 30 min (these were protected from light). Then 50 µL of Stop Solution was added to each well and read spectrophotometrically using Multiskan spectrum spectrophotometer (Thermo Fisher Scientific, Surrey, UK)

at 450 nm within 30min. The coefficient of variation (CV%) for fasting plasma Human C-Reactive Protein was 5%. 6.3.9.5 Calculation of Insulin resistance (IR)/insulin sensitivity (HOMAIR)

The degree of insulin resistance was estimated with the homeostasis model assessment for insulin resistance (HOMA_{IR}) method as a valid surrogate measure of insulin sensitivity. HOMA_{IR} was calculated using a well established formula (Matthews et al., 1985). The product of the fasting concentrations of glucose (expressed as millimol per liter) and insulin (expressed as milliunits per milliliter) is divided by a constant of 22.5 (Equation 6.3).

Equation 6.3:

HOMA_{IR} = <u>fasting glucose (mmol/L) X fasting Insulin (mU/ml)</u> 22.5

6.3.10 Statistical analysis

Comparison of the distribution (means/median) of variables

Due to gender differences in physiology, biochemical profiles and diet, potentially, data were analysed and presented separately by gender. Statistical analyses were performed with IBM SPSS Statistics 21 (IBM Corp., Armonk, NY). Descriptive statistics of the study participants' characteristics (ie, dietary intakes and biochemical measurements) were computed and presented. All variables for which statistical analyses were carried out on were examined for normality (Shapiro Wilks statistics test for normality) and outliers (Box-plot or box-and-whisker plots).

For normally distributed data, Independent Samples T-test (*p*<0.05) was used to compare the averages for variables between group (South Asian and European). For non-normally distributed data, Independent Man-Whitney U test, the non-parametric equivalent, was used. Non-parametric tests are recommended over parametric tests for non-normally distributed data (Vickers, 2005). Mean and standard deviation (SD) were presented for normally distributed variables while median and interquartile range (IQR) or 25th and 75th percentiles of the distribution were presented for non-normally distributed variables.

A subset of males (SA, n=24; EU, n=30) matched for anthropometric (age, body fat %, body mass index) and socio-demographic charactersitics were selected from the main primary data set (SA, n=30; EU, n=30) by selecting only subjects with <23% body fat through IBM SPSS Statistics 21 (IBM Corp., Armonk, NY).

Correlation analysis

The relationship of dietary GI, GL, carbohydrate, SFA, PUFAs and other nutrients respectively, to HOMA_{IR} was tested with Spearman correlation two-tailed test and Spearman Partial correlation test as these were the most appropriate non-parametric correlation tests (Sheskin, 2004) for non-normally distributed data as with the case of most of the tested variables in the present study. Spearman Partial correlation tests were conducted by SPSS syntax script command (IBM, 2014) in IBM SPSS Statistics 21 (IBM Corp., Armonk, NY). Spearman partial correlation tests statistically controlled for variables that correlated significantly with HOMA_{IR} or known confounders (energy intake, body mass index, physical activity level, age, Scottish Index of Multiple Deprivation). Correlation test results with and without statistically controlling for certain variables are presented in the results section.

Stratification of subjects to percentiles of dietary glycaemic index (South Asian males only) and percentile of sugars intake (South Asian females only) following correlation analyses

Following correlation analyses, South Asian males were stratified into lower (n=14) and upper percentile (n=16) group of habitual dietary glycaemic index. Distribution of diet variables and biochemical characteristics were then compared between ethnicity using Independent Samples T-test (p<0.05) for normally distributed data and Independent Man-Whitney U test for non-normally distributed data.

Following correlation analyses, South Asian females were stratified to lower (n=11) and upper percentile (n=11) of sugars intake. Distribution of diet variables and biochemical characteristics were then compared between ethnicity using Independent Samples T-test (p<0.05) for normally distributed data and Independent Man-Whitney U test for non-normally distributed data Stratification of females by sugars intake.

6.4 Results

6.4.1 Males

6.4.1.1 Characteristics of male study subjects

South Asian males

Table 6.1 presents demographic characteristics of male study subjects. Almost all South Asian male subjects (97%) were educated at university level while the remaining obtained up to at least high school education. All South Asians originated from Pakistan of which 40% of them were born in the UK and of British nationality while the remaining were migrants from Pakistan. A majority of them were non-smokers (87%) and an equal number of subjects were married and single. There were also as many employed as unemployed subjects. Slightly over half of the total number of subjects reported a family history of diabetes (57%) while a lower percentage of subjects reported family history of heart problems (23%).

South Asian male subjects were of median age of 29 and their Scottish Index of Multiple Deprivation (SIMD) rank averaged at 3729.97. South Asian males' median body mass index was 23.75 Kg/m² and mean body fat percent was18.95% (SD of 6.33) (Table 6.2). Mean fasting glucose of South Asian male subjects was 4.99mmol/L (SD, 0.40) while their fasting insulin levels averaged to 5.70μ U/L. Their median HOMA_{IR} value and CRP level was1.06 and 0.65mg/L, respectively. They were moderately active with median physical activity level of 1.61 which was derived from self-reported physical activity records.

South Asian and European males

A higher percentage of South Asian males than Europeans were married (53.3% and 10%, respectively) (Table 6.1). The numbers of employed and unemployed subjects were equal in both ethnic groups (50:50). More South Asians than Europeans (37% more) answered "yes" to having family history of diabetes. Among South Asians, a higher percentage of them reported a family history of diabetes (56.7%) than heart problems (23.3%). In Europeans, the number of subjects who reported family history of diabetes and heart problems was similar (approximately 20%).

South Asian and European males were comparable in age, socio-economic status, body mass index (normal weigh category) and waist circumference (Table 6.2). South Asian males, however, had significantly higher body fat (3.67% higher) than European males. There were statistically significant ethnic differences in fasting insulin (*p*-value= 0.037), HOMA_{IR} (*p*-value= 0.024), CRP (*p*-value=0.009) and physical activity expressed as METS-min/week for non-occupational activities such as sports, leisure and way to work (*p*-value=0.000). Fasting insulin of South Asians was 35% higher than that of Europeans while HOMA_{IR} was 16% higher in South Asians. CRP of South Asians was double that of Europeans. South Asians were also 33% less physically active than Europeans in sports and leisure activities. South Asian and European males did not differ significantly in fasting glucose (mean, SD; 4.99, 0.40 and 4.78, 0.43, respectively) nor physical activity level (median of 1.61 and 1.60, respectively).

6.4.1.2 Dietary glycaemic index, glycaemic load and carbohydrate foods in the diet

Dietary glycaemic index, glycaemic load (South Asian males)

Habitual dietary glycaemic index (GI) of South Asian males averaged at 56 (SD of 2.78) and mean glycaemic load (GL) was 156 (SD of 34.27), respectively (Table 6.2). Their median total energy intake was 9011.11KJ/day, equivalent to 2155.77Kcal/day of which energy contributed by carbohydrates was the highest (mean of 48.55%) followed by fat (mean of 35.18%) and protein (median of 14.65%).

Diet characteristics and dietary glycaemic index, glycaemic load (South Asian and European males)

Similarities in dietary characteristics were largely apparent between South Asian and European males only statistically differing in protein intakes (*p*-value= 0.048) (Table 6.2). Total energy intakes were similar at a median of 9011.11KJ/day (equivalent to 2154 Kcal/day) and 10000.79KJ/day (equivalent to 2390 Kcal/day) in South Asian and European males, respectively of which approximately 47% of energy intake was from carbohydrate, 34% from fat and 15% from protein. Median protein intake of South Asians was 17.7g lower than intakes of Europeans per day. Mean habitual dietary glycaemic index of South Asian males (56.20) although higher than European males (54.77), did not reach statistical significance (*p*-value=0.086) and both fell within the medium GI category. Similarly, their habitual dietary glycaemic load was not significantly different (mean, SD; 156.43, 34.27 and 155.20, 47.08). Intakes of carbohydrate, sugars, starch and fibre were also similar between ethnic groups.

For the subset of male subjects (South Asians *n*,24 and Europeans *n*,30) matched for all demographic and anthropometric characteristics including body fat percent as shown in Table 6.3, dietary characteristics were mostly similar to that of the main sample as described above with the exception of ethnic differences in mean habitual dietary glycaemic index which was statistically significant (*p*-value=0.041) but dietary GI of both ethnic groups still fell within medium GI category. Besides protein intakes, starch intakes also significantly differed among this subset of South Asian and European males (*p*-value=0.031). Higher fasting glucose, fasting insulin, HOMA_{IR} and CRP was observed in South Asians than Europeans (only statistically significant for fasting glucose and CRP).

Carbohydrate food intakes (South Asian males)

Main staples in the diet of South Asian males ranked from highest to lowest absolute mean intakes are shown in Table 6.4 ranging from highest mean intake of 163.12 g/day for unleavened breads (chapatti, naan/pitta, paratha) to as low as 2.68 g/day for seeded bread/rye breads and tortillas. Reported intakes of unleavened breads were significantly higher than intakes of other staples in the South Asian diet such as rice, white/wholemeal/brown bread, potatoes, pasta, breakfast cereals and seeded/rye bread and tortilla. Intakes of rice (90.65 g/day) and white/wholemeal/brown bread (62.64 g/day) were comparable. Potatoes, pasta and breakfast cereals were also consumed in similar amounts. Seeded bread/rye bread and tortillas were the least consumed staples in the diet.

Types of breads consumed by South Asian males ranked from most to least consumed was chapatti (no fat), white bread, Naan/Pitta, Pizza, Paratha, the miscellaneous bread food group (burgers, pancakes, crumpets, croissants, fruit loafs and granary roll) and wholemeal bread (Table 6.5). All other breads like English muffins, waffles, buns, baguettes and seeded bread was consumed by a very small number of subjects rendering a value of 0 g/day at quartiles of intakes. South Asian males consumed similar amounts of rice cooked without oil (basmati, long grain and brown rice) and rice cooked with oil like pilau rice and biryani rice (median of 28.72 and 30.71 g/day, respectively). Potatoes were consumed more than pasta and noodles and South Asian males tended to consume more potatoes (chips/fries) that were fried rather than baked. For types of breakfast cereals, high fibre breakfast cereals and lower fibre/sugared cereals were both consumed by South Asian males in similar, albeit in very small amounts.

Other carbohydrate foods in the diet of South Asian males were cakes, biscuits and snacks such as cereal/fruit/chocolate bars and crisps (potato crisps, corn chips). Beverages were also a source of carbohydrates in the diet where soft-drinks, fruit flavoured drinks and cordials were consumed the most while unsweetened fruit juices were consumed in smaller amounts. They also consumed dairy products like milk and yogurt at a median intake of 163.22 g/day, fruits (median of 46.29 g/day) and table sugar/sucrose (9.36 g/day).

Staples such as unleavened breads, rice, bread (white, wholemeal, brown) and potatoes were usually eaten with other foods such as curry, meats, vegetables, soups and butter (Table 6.6). Unleavened breads like chapatti, naan, pitta and paratha were almost universally eaten with curry (chicken/lamb/fish/lentils/potato). Greater amounts of curried chicken/lamb/fish (median of 82.82g/day) tended to be eaten with unleavened breads compared to other types of curry (lentils, potato, vegetables) (median of 41.43 g/day). Rice was eaten with mostly lentil/potato curry. White bread/wholemeal bread tended to be eaten with eggs, cheese/butter or other fat based spreads as well as with meats. Potatoes were usually eaten with meat, vegetables (carrots/corn/peas/leafy vegetables) and fish.

Carbohydrate food intakes (South Asian and European males)

The main staple in the diet of South Asian males was unleavened bread (chapatti, naan/pitta, paratha) while the main staple in the diet of Europeans was white/wholemeal/brown bread (Table 6.4). Both ethnic groups also consumed rice, potatoes, pasta and breakfast cereals but in different quantities. South Asians ate more rice and less bread (white/wholemeal/brown), potatoes, pasta and breakfast cereals than European males.

Amounts of carbohydrate foods consumed (g/day) by South Asian and European males were compared (Table 6.5). For the bread category, South Asian males consumed lesser amounts of wholemeal/brown bread and baguettes (almost significantly different between ethnicity). South Asians consumed wholemeal/brown bread at a 75th PCTL value of 7.78 g/day in contrast to higher intakes by Europeans which was 43.51 g/day at the same percentile. Consumption of all other breads such as white bread, English muffin/waffles/buns, seeded bread/rye bread/tortilla/pizza as well as burgers/pancakes/crumpets etc. were not significantly different between ethnic groups.

For unleavened breads, South Asians consumed significantly more chapatti (no fat), naan/pitta and paratha compared to Europeans. South Asians and European habitually consumed similar amounts of rice cooked without oil or boiled (basmati/long grain/brown rice) but South Asians consumed significantly more rice cooked with oil (Pilau/Biryani) at a median intake of 30.71 g/day (25th,75th PCTL; 0.00, 70.72 g/day) while Europeans consumed none or small amounts of these rice dishes.

South Asian males consumed almost significantly lesser amounts of potatoes (fried/boiled/mashed) (*p*-value=0.069) as well as significantly lesser amounts of pasta (*p*-value=0.001) than European males. Intake of potatoes (baked potatoes/baked chips or fries and boiled sweet potatoes) and noodles was not significantly different between ethnic groups (*p*-value= 0.133 and 0.214, respectively).

For breakfast cereals, South Asian males had lower intakes of low GI, higher fibre varieties like All Bran/Bran Flakes/oatmeal (almost statistically significant, *p*-value=0.051) as well as moderate GI varieties such as Cocoa Crunch/Frosties/Fruit & Fibre/Shredded Wheat etc. (*p*-value=0.002). They had similar intakes of mostly high GI cereals such as Cherrios/Cocoa Pops/Cornflakes/Grapefruits/Puffed Wheat/Ready Brek.

South Asian males had significantly lower intakes of cakes than European males (75th PCTL value of 20.54 g/day and 35.22 g/day, respectively). South Asian males also consumed significantly less amounts of snacks (all types) (*p*-value=0.007) particularly cereal/fruit/protein bars, chocolate bars and potato crisps/corn chips/raisins/marshmallow/popcorn. Intakes of biscuits and crackers as well as snacks such as nuts/trail mix and sweets were similar among males.

Compared to Europeans, South Asians consumed significantly less amounts of beverages (less by 55%) in total (*p*-value= 0.030) especially lemonade/Oat drinks/smoothies/chocolate drink mix/soy protein drink/Ribena (*p*-value=0.011) and soft drinks/fruit flavoured drinks/cordial (almost significant, *p*-value=0.065). Males of both ethnicity drank similar amounts of unsweetened fruit juices and sports drinks. They also had similar intakes of dairy products like milk and yogurt as well as fruits (median intakes of 46.29 g/day and 51.58 g/day in South Asian and European males, respectively).

For amounts of foods consumed with staples (Table 6.6 and 6.7), South Asian males habitually consumed unleavened bread (chapatti/pitta/paratha) with significantly greater quantities of curry, eggs and yogurt, respectively, than European males. South Asian males tended to eat rice with significantly more curry (lentil/potato) and less vegetables (carrot/corn/peas/leafy vegetables). Compared to Europeans, South Asian males consumed white/wholemeal/brown bread with significantly more amounts of curry (chicken/lamb/fish curry and vegetable/lentil/potato curry) and significantly less amounts of meat, vegetables, butter/cheese/oil and soup. In addition, South Asians ate potatoes and pasta, respectively, with significantly less amounts of vegetables. They also consumed potatoes with significantly lesser amounts of butter/cheese/fat-based spreads than Europeans.

6.4.1.3 Correlations of $HOMA_{IR}$ and dietary glycaemic index, glycaemic load and other dietary variables

Table 6.8 shows correlations of HOMA_{IR} with dietary variables. Spearman correlation analysis was carried out on HOMA_{IR} and dietary variables such as habitual dietary GI, dietary GL, carbohydrate, sugars, starch, fibre, protein and fat, respectively. A moderate, statistically significant, negative monotonic relationship between dietary glycaemic index and HOMA_{IR} was observed (r= -0.493, *p*-value=0.006). This relationship persisted, albeit with less significance (r= -0.435 and *p*-value= 0.030) after statistically controlled for total energy intake (KJ/day), body mass index (Kg/m²), age (years), physical activity level and socio-demographic status (SIMD).

Controlling for age particularly lowered the correlation coefficient, r, as well as the *p*-value for the relationship between HOMA_{IR} and dietary GI. Glycaemic load did not relate significantly with HOMA_{IR} (*p*-value= 0.083) and this relationship became weaker when statistically controlled for body mass index and total energy intakes. All other dietary variables were not observed to relate significantly to HOMA_{IR}.

Table 6.9 shows correlations of dietary glycaemic index (GI) to glycaemic load (GL) and GI and GL, respectively, to other dietary variables. With regards to dietary GI, crude Spearman analysis (before controlling for any variables) found that dietary GI related positively to GL and fibre respectively but not to any other diet variables (r=0.467, *p*-value= 0.009 and r=0.425, *p*-value= 0.019, respectively).

Spearman partial correlation analysis (statistically controlled for energy intake, body mass index, age, physical activity level and SIMD) found that the positive relationship between dietary GI and GL as well as dietary GI and fibre persisted and that the strength of the relationship became stronger and more significant (r_c =0.652, *p*-value= 0.000 and r_c =0.510, *p*-value= 0.009, respectively). After controlling for the stated variables, dietary GI was also observed to relate positively to carbohydrate intakes (r=0.500, *p*-value= 0.011) and negatively to

fat intakes (r=-0.475, *p*-value= 0.016). All other dietary variables did not relate significantly to dietary GI.

Dietary GL was found to relate positively to dietary GI as well as carbohydrate, sugars, starch, fibre and total energy intakes and the strength of the relationship ranged from moderate to strong. The relationship between dietary GL and sugars and dietary GL and starch, however, was no longer significant after Spearman Partial Correlation analysis controlling for energy intake, body mass index, age, physical activity level and SIMD. The highest correlation coefficient was observed between dietary GL and carbohydrates with and without controlling for the stated variables above (r=0.972 and r_c =0.931).

Following correlations, South Asian males were stratified into lower and upper percentile of habitual dietary glycaemic index for comparisons of dietary glycaemic index, glycaemic load, carbohydrate related nutrients and HOMA_{IR} (Table 6.10). Males in the lower percentile of dietary GI had significantly lower starch intakes and lower fibre intake than males in the upper percentile of dietary GI. Those in the lower percentile of GI also had higher intakes of dietary fat and fat (%E). All other diet variables did not significantly differ among South Asian males stratified by percentile of GI. Fasting glucose, fasting insulin and HOMA_{IR} was higher for males in the lower percentile of GI (statistically significant for fasting insulin, *p*-value=0.032 and HOMA_{IR}, *p*-value= 0.023) than in the upper percentile of GI. C-reactive protein was similar between the groups.

6.4.2 Females

6.4.2.1 Characteristics of female study subjects

South Asian females

Table 6.12 depicts demographic characteristics of female subjects who participated in the study on dietary glycaemic index, glycaemic load, carbohydrate related nutrients and HOMA_{IR} of South Asians. All South Asian females originated from Pakistan and equal numbers of them were of British nationality (born in the UK) and migrants from Pakistan. Most of them possessed university education (81.8%). The ratio of married to single subjects was also equal. Almost all of them were non-smokers (96%) and a majority of them were unemployed (82%) and this included students.

A majority of them, 77%, reported family history of diabetes which is 46% higher than those who reported family history of heart problems. Their biochemical and physiological characteristics are shown in Table 6.13. The South Asian female subjects were young adults with median body mass index of 25 kg/m² and normal fasting glucose levels (median of 4.70mmol/L). They were moderately active with mean physical activity level of 1.57.

South Asian and European females

A majority of female subjects were non-smokers and a higher percentage of South Asians than Europeans were married (50.0% and 6.9%, respectively). There were more numbers of unemployed than employed subjects in both ethnic groups (18.2% and 37.9% of South Asians and Europeans were employed, respectively). More South Asians than Europeans answered "yes" to having family history of diabetes. Among South Asians, a majority reported a family history of diabetes (77.3%) while a smaller percentage reported family history of heart problems (31.8%). Relatively fewer Europeans reported a family history of diabetes (31%) and heart problems (17.2%).

South Asian and European females were similar in age, socio-economic status, body mass index, body fat percent and waist circumference (Table 6.13). With regards to biochemical and physiological characteristics, there were statistically significant ethnic differences in HOMA_{IR} (*p*-value= 0.037), fasting insulin (*p*- value= 0.029), and physical activity expressed as METS-min/week for sports activities (*p*-value=0.012).

Mean fasting insulin of South Asian females was 38% higher than that of European females. Mean HOMA_{IR} was 36% higher in South Asian than in European females (mean (SD); 1.57 (0.80) and 1.16 (0.58), *p*-value= 0.037, respectively. South Asian and European females did not differ significantly in fasting glucose (median (IQR); 4.70 (0.63) and 4.87 (0.96), respectively). South Asians, compared to their counterparts, had higher CRP levels but this did not reach statistical significance. South Asian females were also significantly less physically active than European females in sports activities, a mean of 0 METSmin/week for South Asians in contrast to 67.50 METS-min/week for Europeans. Physical activity level (PAL) of South Asian females (mean, SD; 1.57, 0.12) was lower than the PAL of European females (mean, SD; 1.66, 0.23) and this almost reached statistical significance (*p*-value=0.052). 6.4.2.2 Dietary glycaemic index, glycaemic load and carbohydrate foods in the diet

Dietary glycaemic index, glycaemic load (South Asian females)

Habitual dietary glycaemic index (GI) of South Asian females fell in the low GI category which was 54 (4.25) and glycaemic load (GL) was 123 (23.75), respectively (values are median (IQR)) (Table 6.13). Their median total energy intake was 7533.83 KJ/day, equivalent to 1800.63 Kcal/day of which mean percent of energy contributed by carbohydrates was the highest (48.59%) followed by fat (mean of 38.50%) and protein (13.21%).

Diet characteristics and dietary glycaemic index, glycaemic load (South Asian and European females)

Similarities in dietary characteristics were largely apparent between South Asian and European females only statistically differing in starch intakes (*p*-value= 0.006) and fat (%E) (*p*-value=0.035) (Table 6.13). Total energy intakes were similar at a mean of 7533.83 KJ/day (equivalent to 1800 Kcal/day) and 7296.50 KJ/day (equivalent to 1744 Kcal/day) in South Asian and European females, respectively of which approximately 49% of energy intake was from carbohydrate and %14 from protein.

Contribution of fat to energy intakes was higher in the diet of South Asians (38.50%) than Europeans (34.21%) and both exceeded the recommendation of 33% (including glycerol) of total dietary energy for the population (Department of Health., 1991). South Asian and European females did not differ in percent of food energy contributed by SFAs and MUFAs. However, compared to European females, South Asian females had significantly more percent energy from PUFAs (7.5%) which exceeded the 6% recommendation for PUFAs while European females were within the DRVs (5.53%) (Table 6.21). The average percent energy from SFA by both groups of females exceeded the recommendation for the UK population of 10% (Department of Health., 1991) of total dietary energy.

For carbohydrate fractions, South Asian females had higher intakes of starch with a mean (SD) of 152.30 (36.09) g/day. Mean habitual dietary glycaemic index of South Asian and European females were similar (median GI of 54) and

within the low GI category. Similarly, their habitual dietary glycaemic load was not significantly different (median (IQR); 123.00, 23.75 and 125.00, 39.00). Intakes of carbohydrates, sugars, fibre, protein and fat were also similar between ethnic groups.

Carbohydrate food intakes (South Asian females)

Main staples in the diet of South Asian females ranked from highest to lowest absolute mean intakes are shown in Table 6.14 ranging from highest mean intake of 84.71 g/day for unleavened breads (chapatti, naan/pitta, paratha) to as low as 19.63 g/day for breakfast cereals. Reported intakes of staples such as unleavened breads, rice, white bread/wholemeal bread/brown bread, potatoes and pasta were not significantly different. Breakfast cereals were taken in significantly lesser amounts than all of these staples and seeded/rye bread and tortilla were not consumed by South Asian females.

Types of breads consumed by South Asian females ranked from most to least consumed was white bread, chapatti (no fat), pizza, naan/pitta, wholemeal bread/brown bread, the bread group -burgers/pancakes/crumpets/croissants/ fruit loafs/granary roll) and paratha (Table 6.15). All other breads like English muffins, waffles, buns, baguettes and seeded bread were consumed by a very small number of subjects or not at all.

South Asian females consumed similar amounts of rice cooked without oil (basmati, long grain and brown rice) and rice cooked with oil like pilau rice and biryani rice (median of 37.32 and 36.43 g/day, respectively). Potatoes and pasta were consumed in comparable quantities while noodles were not prevalent in South Asian females' diet. South Asian females tended to consume more fried/boiled rather than baked potatoes/sweet potatoes.

They mostly consumed lower fibre/sugared breakfast cereal varieties rather than higher fibre varieties. Other carbohydrate foods in the diet of South Asian females were cakes, biscuits and snacks such as cereal/fruit/chocolate bars and crisps (potato crisps, corn chips). Beverages were also a source of carbohydrates in the diet where soft-drinks, fruit flavoured drinks and cordials were consumed the most while unsweetened fruit juices were consumed in smaller amounts. They also consumed dairy products like milk and yogurt at a median intake of 137.29 g/day, fruits (median of 42.86 g/day) and table sugar/sucrose (5.54 g/day).

Staples such as unleavened breads, rice, bread (white, wholemeal, brown) and potatoes were usually eaten with other foods such as curry, meats, vegetables, soups and butter (Table 6.16). Unleavened breads like chapatti, naan, pitta and paratha were almost universally eaten with curry (chicken/lamb/fish/lentils/potato). Higher amounts of curried chicken/lamb/fish (median (IQR) of 61.09 (109.64) g/day) tended to be eaten with unleavened breads compared to other types of curry (lentils, potato, vegetables) (median (IQR) of 24.69 (56.46) g/day).

Rice was mostly eaten with lentil/potato curry followed by curried chicken/lamb/fish. White bread/wholemeal bread tended to be eaten with eggs, vegetables (carrots/corn/peas/leafy vegetables), cheese/butter or other fat based spreads as well as jam/honey/syrup. Potatoes were usually eaten with fish and vegetables (carrots/corn/peas/leafy vegetables).

Carbohydrate food intakes (South Asian and European females)

Main staples in the diet of South Asian females were unleavened bread (chapatti, naan/pitta, paratha), rice, white/wholemeal/brown bread, potatoes and pasta (Table 6.14). European females also consumed these staples in similar amounts for bread, potatoes, pasta and breakfast cereals. Unleavened bread was the exception where Europeans consumed it in small amounts only. South Asian females also consumed more rice.

For the bread category, South Asian females in comparison to European females, consumed significantly more white bread, less wholemeal bread/brown bread, less seeded bread/rye bread/tortilla, more chapatti (no fat) and more paratha. Consumption of all other breads such as English muffin/waffles/buns, baguette/ciabatta/French stick, pizza and burgers/pancakes/crumpets etc. were not significantly different between ethnic groups (Table 6.15).

South Asian females consumed significantly more rice cooked with oil (pilau, biryani) than European females but they ate similar amounts of rice cooked without oil or boiled rice (basmati/long grain/brown rice). Consumption of potatoes, pasta, noodles and breakfast cereals were not significantly different between South Asian and European females.

Females of both ethnicity reported similar intakes of cakes, biscuits/crackers and snacks. South Asian females had lower median intakes of chocolate bars and potato crisps/corn chips/raisins/marshmallow/popcorn than European females but this was just above statistical significance (*p*-value= 0.057).

There were no ethnic differences in consumption of beverages such as lemonade/Oat drinks/smoothies/chocolate drink mix/soy protein drink/Ribena, unsweetened fruit juices, soft drinks/fruit flavoured drinks/cordial and sports drinks. South Asian and European females also had similar intakes of dairy products like milk and yogurt. South Asian females had significantly higher intakes of sugar (sucrose) and lower intakes of fruits compared to European females.

For amounts of foods consumed with staples (Table 6.16 and 6.17), South Asian females habitually consumed unleavened bread (chapatti/pitta/paratha) with significantly more curry and/or yogurt, respectively, than European females. Similarly, rice was also eaten with more curry and yogurt. Europeans, however, tended to consume significantly more vegetables with rice than South Asians.

Compared to Europeans, South Asian females consumed

white/wholemeal/brown bread with significantly more amounts of curry (vegetable/lentil/potato curry) and less amounts of meat and butter/cheese/oil and soup, respectively. South Asian and European females tended to eat potatoes with fish and/or vegetables (carrot/ corn/peas/leafy vegetables) at similar amounts. Pasta tended to be consumed with vegetables (carrot/ corn/peas/leafy vegetables) by both ethnic groups in amounts that were not significantly different. European females also tended to consume pasta with butter/cheese/oil. 6.4.2.3 Correlations of $HOMA_{IR}$ and dietary glycaemic index, glycaemic load and other dietary variables

Results of Spearman correlation analysis for HOMA_{IR} and dietary variables such as habitual dietary GI, dietary GL, carbohydrate, sugars, starch, fibre, protein and fat, respectively for South Asian females is shown in Table 6.18. Crude correlation analysis (not statistically controlled for any potential confounders) did not show any significant relationship between any of the dietary variables and HOMA_{IR} (*p*-value<0.05). A moderate, statistically significant, positive monotonic relationship between sugars intake and HOMA_{IR} was observed (r= 0.486 and *p*-value= 0.048) when statistically controlled for total energy intake (KJ/day), body mass index (Kg/m²), age (years), physical activity level and socio-demographic status (SIMD).

Dietary glycaemic index and glycaemic load did not relate significantly with HOMA_{IR} (*p*-value=0.394 and *p*-value=0.713) statistically controlled for total energy intake, body mass index, age, physical activity level and socio-demographic status (SIMD). All other dietary variables were not observed to relate significantly to HOMA_{IR}. Table 6.19 shows correlations of dietary glycaemic index (GI) to glycaemic load (GL) and GI and GL, respectively, to other dietary variables. With regards to dietary GI, crude Spearman analysis (before controlling for any variables) showed that dietary GI related positively to GL and starch intake respectively but not to any other diet variables (r=0.511, *p*-value= 0.015 and r=0.462, *p*-value= 0.030, respectively). GL related positively to carbohydrates, sugars, starch and total energy intake, respectively.

Spearman partial correlation analysis (statistically controlled for energy intake, body mass index, age, physical activity level and SIMD) showed a persistent positive relationship between dietary GI and GL ($r_c=0.505$, p-value= 0.039) as well as dietary GL and carbohydrate and starch, respectively ($r_c=0.914$, p-value= 0.000 and $r_c=0.563$, p-value= 0.019) but relationship between GL and sugars was no longer significant (p-value=0.05). An inverse relationship between dietary GL and fat intake became significant ($r_c=-0.750$, p-value= 0.001) in partial spearman analysis. Sugars intake related positively to carbohydrate intakes and negatively to protein intake ($r_c=0.509$, *p*-value= 0.037 and $r_c=-0.545$, *p*-value= 0.024, respectively)

Following correlations, South Asian females were stratified into lower and upper percentile of sugars intake for comparison of dietary glycaemic index, glycaemic load, carbohydrate related nutrients and HOMA_{IR} (Table 6.20). Females in the upper percentile of sugars intake had significantly higher habitual dietary glycaemic load and higher intakes of total carbohydrate. All other diet variables did not significantly differ among females in the lower and upper percentile groups of sugar intake. Fasting glucose, fasting insulin and HOMA_{IR} of females in the upper percentile of sugars intake was higher than that of the lower percentile of sugars intake and this difference was statistically significant for fasting glucose that was approximately 8% higher than fasting glucose of those in the lower percentile of sugars intake. C-reactive protein was higher among females in the lower percentile of sugars intake. (C-reactive protein was higher among females in the lower percentile of sugars intake) 8% higher than fasting glucose of those in the lower percentile of sugars intake. C-reactive protein was higher among females in the lower percentile of sugars intake but this was not statistically significantly higher (*p*-value=0.974).

<u>Correlation of HOMA_{IR} with dietary saturated fatty acids, polyunsaturated fatty</u> <u>acids and monounsaturated fatty acids in South Asians</u>

In South Asians (males and females, n=52), percent energy from total dietary fat intake, saturated fatty acids (SFAs), polyunsaturated fatty acids (PUFAs) and monounsaturated fatty acids (MUFAs) were not found to relate to HOMA_{IR} (Table 6.21). When stratified into gender, PUFAs intake was found to inversely relate to HOMA_{IR} in South Asian females only (Table 6.22).

6.4.3 Study compliance

A total of 142 subjects were initially recruited for this study. Of this total, 111 (78%) subjects completed the study and provided complete data that were deemed valid and further analyzed. They comprised of 60 males (30 South Asians and 30 Europeans) and 51 females (22 South Asians and 29 Europeans). The total study drop-out rate was 22% (31 out of 142). Drop-out rate was higher among females (27%) than males (17%). By ethnicity, drop-out out rate for South Asians (25%) was higher than for Europeans (19%). The drop-out rate for South Asian females included those who were withdrawn from the study because it was later known that they no longer fulfilled study criteria (pregnancy). Subjects who dropped out or were withdrawn from the study did not provide complete data for analysis and were excluded from further data analysis.

The number of days weighed food intake records and Food Frequency Questionnaire were collected and the percentage of subjects who completed those records in the given days are as follows: South Asian males (n=30), 5-7 days, 77%; 3-4 days, 23% European males (n=30), 5-7 days, 87%; 3-4 days, 13% South Asian females (n=22), 5-7 days, 68%; 3-4 days, 32% European females (n=29), 5-7 days, 76%; 3-4 days, 24%

6.5 Discussion

Higher HOMA_{IR} in South Asians than Europeans

This current study confirms that South Asians are more insulin resistant than Europeans. This has been widely documented in the past (Forouhi et al., 2006;Lovegrove, 2007;Merchant et al., 2007;Mente et al., 2010;Ghouri et al., 2013;Goff et al., 2013;Sleddering et al., 2014). Furthermore, fasting insulin and C-reactive protein (in males only) was higher among South Asians than Europeans and this has also been observed previously (Ghouri et al., 2013). South Asians in the current study were found to be less physically active in sports activities particularly women and many studies have made these same observations (Hayes et al., 2002;Williams et al., 2011a). South Asians have also been found to have lower cardiorespiratory fitness compared to matched Europeans (Ghouri et al., 2013).

Habitual Dietary GI and GL

The diets of South Asians to Europeans were compared to identify if there were any differences that could explain for the higher HOMA_{IR} observed in South Asians. Habitual dietary GI and GL did not differ significantly between South Asians and Europeans. Although mean dietary GI was found to be in the medium GI category for South Asian females while dietary GI of European females fell in the low GI category; their mean dietary GI did not differ significantly. The mean dietary GI of South Asian and European males fell in the medium GI category and were also not significantly different. This was similar to the study by Goff et al., (2013) which reported that the GI of both males and females of South Asian and European ethnicity were in the medium GI category but whether dietary GI of males and females significantly differed by ethnicity in that study is unknown because statistical comparison of dietary GI and GL in that study included 3 ethnic groups (South Asians, Europeans and Black-African Carribeans).

In this current study, South Asians probably elicit a medium glycaemic response on average. South Asians and Europeans did not differ in their dietary GI and GL implying that dietary GI and GL per se probably do not explain for higher insulin resistance among South Asians. It is however, important to note that perhaps it is not only average "dietary GI value" that matters but how individuals metabolically respond to the whole diet at that particular dietary GI. South Asians and Europeans have been shown to differ in their glycaemic response (Henry et al., 2008) where glycaemic response of South Asians tended to be higher than that of Europeans when they tested the same foods. Therefore at the same dietary GI, South Asians would probably exhibit higher glycaemic response which could cause higher insulinemic response that over time, could increase fasting glucose and/or fasting insulin which could eventually lead to higher HOMA_{IR} and insulin resistance in South Asians.

In South Asian females, dietary GI and GL did not relate to HOMA_{IR}. This is probably because the diet on average, were medium in GI and the GI range was not very wide due to little variation in the diet in terms of carbohydrate food types (and hence, dietary GI) as well as in the way carbohydrate foods/staples were consumed whether alone or in combination with other foods (mixedmeals). We observed that South Asians tended to eat their foods in similar combinations. Chapatti and rice, for instance was universally eaten with some type of curry (chicken, lamb, lentils or vegetables). Another reason could be due to dietary fat intake. It is clear that co-ingestion of carbohydrate foods with other foods containing protein and fat lower the glycaemic response to the meal (Henry et al., 2006; Moghaddam et al., 2006; Dodd et al., 2011). So when the fat intake is relatively high, as the in the case of South Asian females (38.5% energy) from fat), the dietary GI will tend to be low on average because a fair amount of foods in their diet are probably high in fat which has a glycaemic lowering effect. Rice/chapatti and curry meals, depending on the type of curry whether vegetable, chicken, lamb or potato based, and the amount of oil added in cooking, contained from as low as 28% to as high as 45% energy from fat. A bigger sample size would probably give a wider range of dietary GI (assuming that the amount of fat intakes in the population varies) from which the hypothesis for dietary GI and HOMA can be tested more rigorously.

Dietary Fat and other diet variables

South Asian females were observed to have considerably higher intakes of fat (38.5% total dietary energy) compared to their counterparts (34.21% total

dietary energy). We also observed that South Asian females had higher starch intake than European females (*p*-value, 0.006) and that South Asian males has significantly less intakes of protein than European males.

The percentage of fat consumed by South Asians females (38.5% of food energy) was well above the recommended fat intake of 33% of total energy per day for the general population and remarkably similar to the figures reported in previous studies for South Asians which was 39.9% in British born SA females and 42.4% in migrant SA females (Anderson and Lean, 2005); 35% and 38% for vegetarian and non-vegetarian males and females (Joshi and Lamb, 2000), respectively and 38% in SA females (Reddy and Sanders, 1992). High intake of dietary fat is known to contribute to the development of insulin resistance (Lovejoy et al., 2001;Wang et al., 2003;Ralston et al., 2013). A high plasma free fatty acids (FFA) concentration is associated with an increased risk of developing diabetes (Paolisso et al., 1995) possibly by reducing insulin secretion (Zhou and Grill, 1994;Carpentier et al., 1999) and insulin action (Boden et al., 1994) which are factors involved in development of diabetes Type 2 (Ferrannini, 1998).

In this study, saturated fatty acid (SFA) intakes did not differ between SA and EU but SFA intakes were above the recommended level of 10% of total dietary energy for the UK (Department of Health., 1991) in all groups, the highest being in SA females. Overall consumption of dietary SFAs has been shown to positively relate to insulin resistance (Marshall et al., 1997;Manco et al., 2000;Riserus et al., 2007;Isharwal et al., 2009), fasting insulin and postprandial insulin levels (Parker et al., 1993). Recently diagnosed diabetics as well as subjects with undiagnosed type 2 diabetes had significantly higher consumption of saturated fat compared with controls (Thanopoulou et al., 2003;van de Laar et al., 2004).

Furthermore, higher intakes of SFAs compared to MUFAs have been associated with decreased insulin sensitivity (Galgani et al., 2008;Lopez et al., 2011). In this study however, South Asian females' intake of monounsaturated fatty acids was higher than saturated fatty acid intake (p-value=0.027). Therefore higher HOMA_{IR} in South Asians was not likely due to higher intakes of saturated fats in comparison to monounsaturated fats.
Although SFA intakes did not differ between SA and EU in this current study, South Asians may be more sensitive to the detrimental impact of a high SFA diet compared to Europeans because South Asians are predisposed to insulin resistance and diabetes Type 2 (Bhopal et al., 2014). High SFA diet as opposed to high MUFA diet, have been shown to adversely impact postprandial glycaemic function in subjects predisposed to insulin resistance such as individuals with normal and high fasting triglycerides (Bermudez et al., 2014), in obese and Type 2 diabetics (Christiansen et al., 1997;Summers et al., 2002), obese and insulin resistant (Paniagua et al., 2007) and healthy moderately overweight individuals (Paniagua et al., 2007). Individuals with enhanced risk of type 2 diabetes, daughters of Type 2 diabetic patients for instance, are more sensitive to high intake of SFA in comparison to individuals with lower risks of type 2 diabetes ie. in women with no family history of diabetes (Ntali et al., 2010).

However, in this study, the percent energy (%E) from SFAs, MUFAs and PUFAs respectively of South Asians (known to be predisposed to type 2 diabetes), did not relate to HOMA_{IR} (males and females combined, n= 52) (Table 6.21). This may be because the range of SFA, MUFA and PUFA intake fell within a small range as South Asians had mostly high intakes of total dietary fat. Although %E from SFAs did not relate to HOMA_{IR}, SFA intakes were above 10% and this may have a detrimental impact on insulin sensitivity of South Asians as discussed previously.

When South Asians were stratified by gender, PUFAs was found to relate inversely with HOMA_{IR} (r, -0.510; p, 0.037) in South Asians females (Table 6.22). Previous studies have also shown that intakes of PUFAs inversely relate with HOMA_{IR} (Borkman et al., 1993;Isharwal et al., 2008). Therefore a lower intake of PUFAs in South Asians than in Europeans may explain for the higher HOMA_{IR} in South Asians but this is not the case in the current study because South Asians had significantly higher intake of PUFAs compared to Europeans. Perhaps the types of dietary PUFAs intakes explain for these differences or South Asians may differ in their sensitivity to different types and amounts of dietary fats compared to Europeans. South Asians (males and females) were found to have higher intakes of PUFAs in their diet compared to Europeans. The ratio of higher dietary n-6 PUFAs to lower n-3 PUFAs have been linked with insulin resistance among South Asians (Lovegrove, 2007). A limitation in this study is that the dietary analysis software (WinDiets) only provided the total dietary PUFAs but did not provide the breakdown of n-6 PUFAs and n-3 PUFAs. In this study, however, the higher PUFAs in the South Asian diet was probably in the form of mostly n-6 PUFA sourced from mostly vegetable oils and n-3 PUFAs to a smaller extent (South Asian subjects did not prevalently take foods rich in n-3 PUFAs like fish and fish oils). Previous studies have also observed higher PUFA intakes among South Asians and suggested that the increased PUFA is in the form of n-6 PUFA from vegetables oils (Lovegrove, 2007) in combination with a lower intake of n-3 PUFA (which is cardioprotective) (Sevak et al., 1994;Lovegrove, 2007). The lower dietary intake of LC n-3 PUFAs alone, or in combination with the high n-6 PUFA intake has been suggested to explain for the low LC n-3 PUFA status reported in Indian Asian groups and these profiles have been implicated in insulin resistance (Lovegrove, 2007). In contrast to this, long chained PUFAs have been shown to positively relate to insulin sensitivity (Borkman et al., 1993; Isharwal et al., 2008).

Relationship of dietary variables to HOMA_{IR} (South Asians)

For males, fat intake was similar between the ethnic groups but correlation analyses revealed that he lower the dietary GI, the higher the HOMA_{IR}. This seemed paradoxical at first because it is against existing hypothesis of "the higher the GI, the higher the HOMA_{IR} /risk of diabetes/risk of CHD". This relationship, however, was explained by the observation that GI was inversely related to fat intakes and positively related to fiber intakes. This means that a low GI diet was also high in fat and low in fiber. It is this "low GI, high fat, low fiber" diet pattern that related to HOMA_{IR} and not "low dietary GI" alone. Dietary GI was able to capture this pattern that related to insulin resistance.

In South Asian females, dietary GI did not relate to HOMA_{IR} but HOMA_{IR} was found to increase with sugars intake. Although total sugars intake did not differ among South Asians and Europeans, South Asians consumed more sucrose (table

sugar) which was mainly added to beverages like tea and coffee. Two metaanalysis and systematic reviews have concluded an association between sugarsweetened soft drink intake and type 2 diabetes risks (Malik et al., 2010;Greenwood et al., 2014). Sugar sweetened beverages have been shown to raise blood glucose and insulin concentrations rapidly and dramatically (Janssens et al., 1999) and usually consumed in large amounts which contribute to high dietary glycemic load. High glycemic load diets are known to induce glucose intolerance and insulin resistance among overweight individuals (Schulze et al., 2004) and can increase levels of inflammatory biomarkers such as C-reactive protein, which are associated to type 2 diabetes risk (Liu et al., 2002). However, this positive relationship between sugars intake and HOMA_{IR} must be interpreted with caution because the overall diet must be considered.

When South Asian females were stratified into lower and upper percentile of sugars intake (Table 6.20), South Asian females that were in the upper percentile of sugars intake had high fat intakes (37%) and their mean GI was in the medium GI category. Those in the upper percentile of sugars intake had higher fasting glucose, fasting insulin and $HOMA_{IR}$ (although only statistically significant for fasting glucose). Based on this, the diet of South Asian females that were medium GI, high in fat and high in sugars may contribute to a higher HOMA_{IR} in South Asian females. It was a "medium GI, high fat, high sugars diet" that probably attributed to insulin resistance in South Asian females. This diet tended to be high in carbohydrate as well because sugar is a fraction of carbohydrates and GL is a product of carbohydrate and GI. Although sugars intake did not relate to fat intake in correlation analysis, the diet was high in fat on average. Fat probably did not relate to HOMA_{IR} in correlation analyses because a majority of South Asian females had high intakes of fat (86% of South Asian females had >33% fat from total dietary energy, %fat energy ranged from 29% to 47%).

Therefore it was this "medium GI, high fat, high sugars diet" pattern as opposed to the diet pattern of European females which tended to be "low GI, relatively low fat diet" that explains for the higher $HOMA_{IR}$ among South Asian females. Indeed dietary patterns have been shown to contribute to cardiovascular disease (CVD) risk in South Asians (Gadgil et al., 2014) where a

"vegetarian diet", compared to the "western diet" among Asian Indians living in the United States was associated with lower homeostasis model of assessmentinsulin resistance and lower high-density lipoprotein cholesterol. Both dietary patterns were associated with adverse metabolic outcomes so the authors concluded that healthy food/diet choices could help Asian Indians improve risk factors for CVD (Gadgil et al., 2014).

These findings suggest that genetic predisposition (of South Asian ethnicity, higher reported cases of family history of diabetes and lower cardiorespiratory health), having a dietary pattern as mentioned above, as well as being physically less active in sports activities all together explain for the higher insulin resistance among South Asians. In a high fat diet, the dietary GI and GL probably do not have an impact on insulin resistance but rather it is the interaction of fat and other diet variables such as sugars intake and fibre that may affect insulin resistance. Insulin resistance promoting factors may include a combination of a diet high in fat, higher in carbohydrate (sugars) and lower in fibre in South Asians. These factors together, along with being less physically active especially in vigorous activities, may promote insulin resistance especially in South Asians who are genetically predisposed to chronic disease.

Extrapolation of the results of this study to the population of South Asians in Glasgow is limited because the study did not include a random sample of the population. The population studied probably had higher education and income than average for the population of Glasgow because a fair number of them were highly educated (university level). Nevertheless, the samples of the South Asian and white European populations obtained were fairly well matched for anthropometric and demographic characteristics and the objectives of the study were achieved. Most importantly, this study was a hypothesis-generating study and requires future confirmatory studies.

Bearing in mind that these data were from an observational study, causality cannot be concluded. However, these findings support current healthy eating and diet advice to avoid excess intakes of dietary fat and to eat a well-balanced diet containing sufficient fruits and vegetables (fibre). These research findings could also provide further important evidence that public health initiatives and interventions should not focus solely on the diet but also give special emphasis on promoting increase of physical activity, particularly vigorous activities like sports and exercise, for the health and well-being of South Asian populations.

6.6 Conclusion

Habitual dietary GI and GL did not differ significantly between South Asians and Europeans therefore dietary GI and GL alone possibly do not explain for the higher insulin resistance among South Asians compared to Europeans. In individuals predisposed to insulin resistance like South Asians, a medium GI diet that is high in fat (particularly in saturated fat), low in fibre and high in sugars together with low physical activity namely in structured exercise like sports may explain for insulin resistance.

Table 6.1: Demographic characteristics of male study subjects

South Asian males (*n* 30) European males (*n* 30)

Variable

	Prevalence (%)	Prevalence (%)
¹ Highest education level (university)	96.7	80.0
² British nationality (born in the UK)	40.0	
³ Non-smokers	86.7	83.3
Married	53.3	10.0
Employed	50.0	53.3
Family history of diabetes (answered "yes")	56.7	20.0
Family history of heart problems (answered "yes")	23.3	23.3

¹ the remaining percentage of subjects obtained up to high school level education

² remaining South Asian males were migrants from Pakistan

³ for South Asian males; unknown/did not answer= 3.3%

Table 6.2: Dietary glycaemic index, glycaemic load, carbohydrate-related nutrients and $HOMA_{IR}$ of South Asian and European males

Variable		[†] <i>p</i> -value									
	South As	South Asian males (<i>n</i> 30)		an males 30)							
Demographic and anthropon Age (years)	netric chara 29.00	cteristics (7.00)	26.00	(10.50)	(0.276)						
SIMD	3729.97,	1288.56	3297.77,	1764.93	0.283						
BMI (kg/m ²)	23.75	(3.70)	23.05	(3.55)	(0.290)						
Body Fat %	18.95,	6.33	15.28,	3.94	0.009						
Waist circumference (cm)	83.20	(14.91)	80.65	(7.55)	(0.446)						
Dietary characteristics											
Glycaemic index	56.20,	2.78	54.77,	3.53	0.086						
Glycaemic load	156.43,	34.27	155.20,	47.08	0.908						
Carbohydrate (g/day)	278.46,	53.92	282.33,	80.94	0.828						
Sugars (g/day)	74.37	(43.40)	98.80	(57.53)	(0.174)						
Starch (g/day)	168.75	(39.48)	161.03	(54.87)	(0.198)						
Fibre (g/day)	18.51	(8.67)	17.51	(7.31)	(0.228)						
Protein (g/day)	77.45	(30.56)	95.17	(21.75)	(0.048)						
Fat (g/day)	83.82	(20.09)	92.11	(36.67)	0.535						
Carbohydrate (%E)	48.55,	6.30	46.09,	7.75	0.182						
Protein (%E)	14.65	(4.90)	15.90	(4.40)	(0.145)						
Fat (%E)	35.18,	4.17	33.47,	5.51	0.178						
Total energy intake (KJ/day)	9011.11	(1542.34)	10000.79	(3822.46)	(0.274)						
Biochemical and physiologic	al characte	ristics									
Fasting glucose (mmol/L)	4.99,	0.40	4.78,	0.43	0.063						
Fasting insulin (µU/L)	5.70,	3.50	4.21,	1.55	0.037						
HOMA _{IR}	1.06	(0.58)	0.91	(0.47)	(0.024)						
CRP (mg/L)	0.65	(1.28)	0.28	(0.53)	(0.009)						
¹ Physical Activity Level	1.61	(0.34)	1.60	(0.36)	(0.879)						
METS-min/week for sports, leisure and way to work	942.22,	364.29	1403.27,	507.38	0.000						

n, sample size; SD, standard deviation; IQR, interquartile range; SIMD, Scottish Index of Multiple Deprivation rank score; E, energy intake; HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance;

¹calculated from subjects' self-reported physical activity records;

[†]*p*-value not in brackets was calculated by Independent Samples T-Test for normally distributed variables; *p*-value in brackets was calculated by Man-Whitney U test for non-normally distributed variables;

Statistical significance was set at p < 0.05

Table 6.3: Dietary glycaemic index, glycaemic load, carbohydrate-related nutrients and HOMA_{IR} of South Asian and European males (subset of SA and EU males matched for demographic and anthropometric characteristics)

Variable	I	Mean, SD or	Median (IQF	R)	[†] <i>p</i> -value					
	South Asian males (<i>n</i> 24)		Europea (<i>n</i>	an males 30)						
Demographic and anthropon Age (years)	etric chara 28.00	cteristics (7.50)	26.00	(10.50)	(0.524)					
SIMD	3690 13	1366 75	3297 77	1764 93	0.375					
BMI (kg/m ²)	23.00	(3.30)	23.05	(3.55)	(0.993)					
Body Fat %	16.76,	4.27	15.28,	3.94	0.193					
Waist circumference (cm)	82.30	(8.10)	80.65	(7.55)	(0.903)					
Dietary characteristics										
Glycaemic index	56.63,	2.83	54.77,	3.53	0.041					
Glycaemic load	163.25,	32.04	155.20,	47.08	0.478					
Carbohydrate (g/day)	288.47,	51.23	282.33,	80.94	0.748					
Sugars (g/day)	87.51,	34.52	102.34,	50.69	0.227					
Starch (g/day)	188.76,	51.45	161.38,	41.85	0.036					
Fibre (g/day)	19.64	(9.03)	17.51	(7.31)	(0.141)					
Protein (g/day)	77.45	(26.61)	95.17	(21.75)	(0.031)					
Fat (g/day)	85.84	(24.20)	92.11	(36.67)	(0.689)					
Carbohydrate (%E)	49.19,	5.43	46.09,	7.75	0.103					
Protein (%E)	14.30	(2.80)	15.90	(4.40)	(0.062)					
Fat (%E)	34.98,	4.01	33.47,	5.51	0.266					
Total energy intake (KJ/day)	9362.75	(1695.01)	10000.79	(3822.46)	(0.465)					
Biochemical and physiologic	al characte	ristics								
Fasting glucose (mmol/L)	4.91	(0.57)	4.70	(0.52)	(0.030)					
Fasting insulin (µU/L)	4.69,	1.56	4.21,	1.55	0.268					
HOMA _{IR}	1.05,	0.38	0.90,	0.36	0.132					
CRP (mg/L)	0.55	(1.23)	0.28	(0.53)	(0.022)					
¹ Physical Activity Level	1.62	(0.36)	1.60	(0.36)	(0.454)					
METS-min/week for sports, leisure and way to work	962.73,	395.67	1403.27,	507.38	0.001					

n, sample size; SD, standard deviation; IQR, interquartile range; SIMD, Scottish Index of Multiple Deprivation rank score; E, energy intake; HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance; METS, metabolic equivalent task score

¹calculated from subjects' self-reported physical activity records;

[†]*p*-value not in brackets was calculated by Independent Samples T-Test for normally distributed variables; *p*-value in brackets was calculated by Man-Whitney U test for non-normally distributed variables;

Statistical significance was set at p -value <0.05

Staples intake derived from	weighed food	d records for	males (g/da	у)
	S	outh Asian	Europ	ean
		(<i>n</i> 30)	(<i>n</i> 3	0)
	Mean	SD	Mean	SD
Unleavened bread	163.12 ^ª	124.20	6.44 ^a	14.27
(Chapatti, Naan/Pitta, Paratha)				
Rice (all types)	90.65 ^b	97.81	46.41 ^a	51.61
Bread	62.64 ^b	43.86	115.36 [⊳]	77.60
(White, Wholemeal, Brown)				
Potatoes	32.41 [°]	40.71	58.95 ^b	63.00
Pasta	25.07 ^c	43.92	78.49 ^b	73.85
Breakfast Cereals	16.83 [°]	38.12	43.96 ^a	58.20
Bread (Seeded, Rye, Tortilla)	2.68 ^d	11.48	2.64 ^{ac}	8.37

Table 6.4: Staples intake for males by ethnicity

Different superscript letters indicate significant difference in distributions of staples in the same column (analysed with Friedman's pairwise comparisons test; significance level was set at p < 0.05)

Food type	Amount consumed by males (g/day)						
	South Asian (<i>n</i> 30)			Eur	opean (<i>n</i>	30)	
	Median	25 th PCTL	75 th PCTL	Median	25 th PCTL	75 th PCTL	<i>p</i> -value
Breads							
White bread	54.09	17.83	92.84	79.79	23.18	146.93	0.137
Wholemeal bread, Brown bread	0.00	0.00	7.78	3.86	0.00	43.51	0.058
English Muffin, Waffle, Bun (Chelsea)	0.00	0.00	0.00	0.00	0.00	0.00	0.966
Baguette, Ciabatta, French Stick	0.00	0.00	0.00	0.00	0.00	2.50	0.057
Seeded bread, Rye bread, Tortilla	0.00	0.00	0.00	0.00	0.00	0.00	0.996
Pizza	0.00	0.00	46.43	0.00	0.00	52.86	0.954
Burger, pancake, crumpet, croissant, fruit loaf,	10.86	0.00	35.84	16.48	0.00	56.43	0.281
granary roll							
Unleavened bread							
Chapatti (no fat)	57.57	7.50	152.50	0.00	0.00	0.00	0.000
Chapatti (with fat)	0.00	0.00	0.00	0.00	0.00	0.00	0.317
Naan/Pitta	6.79	0.00	55.54	0.00	0.00	0.00	0.003
Paratha	0.00	0.00	36.19	0.00	0.00	0.00	0.000
Rice							
Rice cooked without oil (Basmati, long grain, brown rice)	28.72	0.00	60.00	23.36	0.00	70.77	0.660
Rice cooked with oil (Pilau, Biryani)	30.71	0.00	70.72	0.00	0.00	0.00	0.000

Table 6.5: Amount of carbohydrate foods consumed (g/day) by South Asian (n 30) and European (n 30) males

PCTL, percentile

P-value for Man-Whitney U test, Statistical significance was set at p < 0.05

Food type							
	Sout	th Asian (n 30)	Eur	opean (<i>n</i>	30)	
	Median	25 th PCTL	75 th PCTL	Median	25 th PCTL	75 th PCTL	<i>p</i> -value
Potato							
Potato- fried (chips/fries), boiled, mashed	14.07	0.00	57.26	29.81	16.00	65.94	0.069
Potato- baked (chips/fries), boiled sweet potato	0.00	0.00	0.00	0.00	0.00	0.00	0.133
Pasta	0.00	0.00	46.57	68.99	0.00	131.93	0.001
Noodles	0.00	0.00	2.50	0.00	0.00	0.00	0.214
Breakfast cereal							
All Bran, Bran flakes, Oatmeal	0.00	0.00	0.00	0.00	0.00	2.25	0.051
Cocoa Crunch, Frosties, Fruit & Fibre, Shredded Wheat, Special K, Weetabix, Porridge, Muesli	0.00	0.00	6.75	19.06	0.00	46.62	0.002
Cherrios, Cocoa Pops, Cornflakes, Grapefruits, Puffed Wheat, Ready Brek, Rice Krispies, Rice Pops, Shreddies, Sugar Puffs, Weetos	0.00	0.00	9.29	0.00	0.00	12.95	0.944

P-value for Man-Whitney U test, Statistical significance was set at p<0.05

Food type							
	Sout	h Asian (<i>i</i>	n 30)	European (<i>n</i> 30)			
	Median	25 th PCTL	75 th PCTL	Median	25 th PCTL	75 th PCTL	<i>p</i> -value
Cakes	0.00	0.00	20.54	17.64	0.00	35.22	0.047
Biscuits and crackers							
Riscuits (all types)	8.47	0.00	25.50	7.92	0.00	16.34	0.767
Digestives, Grain crackers, Oat biscuits, Rich Tea biscuits, Ryvita	0.00	0.00	7.95	0.00	0.00	10.30	0.586
Cream Crackers, Crispbread Rye, Oatcakes, Cream filled biscuits, Shortbread biscuits	0.00	0.00	3.43	0.00	0.00	0.00	0.213
Rusks, Wafer biscuits, Pop tarts	0.00	0.00	0.00	0.00	0.00	0.00	0.127
Snacks							
Snacks (all types)	10.57	0.00	33.13	38.84	11.65	63.86	0.007
Cereal/fruit/protein bars, Chocolate bars	0.00	0.00	12.72	7.40	0.00	25.00	0.080
Nuts, trail mix	0.00	0.00	1.44	0.00	0.00	10.71	0.199
Potato crisps, Corn chips, Raisins, Marshmallow, Popcorn	0.00	0.00	10.43	10.50	0.00	28.21	0.002
Sweets, fruit gums/Jelly, Jelly beans	0.00	0.00	0.00	0.00	0.00	6.79	0.403

P-value for Man-Whitney U test, Statistical significance was set at p < 0.05

Food type	Amount consumed by males (g/day)						
	Sout	n Asian (<i>i</i>	n 30)	Euro	opean (<i>n</i>	30)	
	Median	25 th PCTL	75 th PCTL	Median	25 th PCTL	75 th PCTL	<i>p</i> -value
Beverages							
Beverage (all types)	156.25	28.57	339.87	346.93	99.79	483.46	0.030
Lemonade,Oat drinks, Smoothies, Chocolate drink mix, Soy protein drink, Ribena	0.00	0.00	0.00	0.00	0.00	89.39	0.011
Unsweetened fruit juices	26.79	0.00	99.65	0.00	0.00	77.14	0.660
Soft drinks, fruit flavoured drinks, cordials	81.52	0.00	221.07	155.14	66.32	381.63	0.065
Sports drinks	0.00	0.00	0.00	0.00	0.00	0.00	0.652
Dairy (milk, yogurt)	163.22	88.40	250.18	152.79	81.50	252.40	0.935
Sugar (Sucrose)	9.36	2.97	14.50	0.00	0.00	1.07	0.000
Fruits	46.29	8.44	110.25	51.58	23.52	153.22	0.373

P-value for Man-Whitney U test, Statistical significance was set at *p*<0.05

Table 6.6: Amount of foods consumed	(g/day)	with staples b	y South Asian	males (<i>n</i> 30)
	VU V			· · · · · · · · · · · · · · · · · · ·

	Amount of foods consumed with staples (g/day)										
Foods eaten with staples	Chapatt Pitta/P	i/Naan/ aratha	/ Rice		Bread		Potatoes (not including potatoes in curry)		Pasta		
	Median	IQR	Median	IQR	Median	IQR	Median	IQR	Median	IQR	
^{1,3} Curry (chicken/lamb/fish)	82.82	106.25	0.00	30.72	0.00	0.00	0.00	0.00	0.00	0.00	
^{1,2,3} Curry (lentils, potato)	41.43	86.96	6.43	28.93	0.00	0.00	0.00	0.00	0.00	0.00	
³ Meat	0.00	2.50	0.00	1.32	0.00	16.77	0.00	18.47	0.00	0.00	
Fish	0.00	0.00	0.00	0.00	0.00	2.43	0.00	13.22	0.00	0.00	
¹ Eggs	0.00	9.28	0.00	0.00	4.07	23.96	0.00	0.00	0.00	0.00	
Beans and legumes	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
^{2,3,4,5} Carrot/ corn/peas/leafy vegetables	0.00	2.86	0.00	0.00	0.00	1.86	0.00	0.00	0.00	0.00	
Pumpkin, turnips, swede	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
^{3,5} Butter/ cheese/oil	0.00	0.00	0.00	0.00	1.43	10.14	0.00	0.00	0.00	0.00	
Jam/honey/syrup	0.00	0.00	0.00	0.00	0.00	4.07	0.00	0.00	0.00	0.00	
¹ Yogurt	0.00	1.07	0.00	2.32	0.00	0.00	0.00	0.00	0.00	0.00	
³ Soup	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	

IQR, Interquartile range

Quantity (grams/day) consumed with ¹Chapatti/Naan/pitta/Paratha, ²rice, ³bread (white/wholemeal/brown) ⁴potatoes, ⁵pasta, respectively, were significantly different between South Asian and European males (*p*<0.05) when tested using Man-Whitney U test

Table 6.7: Amount of foods consumed (g/day) with staples by European males (n 30)

	Amount of foods consumed with staples (g/day)										
Foods eaten with staples	Chapatti Pitta/Pa	/Naan/ Iratha	Rice		Bread		Potatoes (not including potatoes in curry)		Pasta		
	Median	IQR	Median	IQR	Median	IQR	Median	IQR	Median	IQR	
Curry (chicken/lamb/fish)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
Curry (lentils, potato)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
Meat	0.00	0.00	0.00	30.77	38.02	50.82	6.07	37.87	0.00	0.00	
Fish	0.00	0.00	0.00	0.00	0.00	0.00	0.00	25.00	0.00	0.00	
Eggs	0.00	0.00	0.00	0.00	0.00	8.04	0.00	0.00	0.00	0.00	
Beans and legumes	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
Carrot/ corn/peas/leafy vegetables	0.00	0.00	0.00	14.43	9.40	23.90	4.79	27.73	0.00	2.05	
Pumpkin, turnips, swede	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
Butter/ cheese/oil	0.00	0.00	0.00	0.00	9.79	21.55	0.00	5.71	0.00	0.00	
Jam/honey/syrup	0.00	0.00	0.00	0.00	0.00	7.13	0.00	0.00	0.00	0.00	
Yogurt	0.00	0.00	0.00	0.42	0.00	0.00	0.00	0.00	0.00	0.00	
Soup	0.00	0.00	0.00	0.00	0.00	44.19	0.00	0.00	0.00	0.00	

	South Asian males (<i>n</i> 30) HOMA _{IR}									
Variables	r	r _{c1}	r _{c2}	r _{c3}	r _{c4}					
	(p-value)	(p-value)	(p-value)	(p-value)	(p-value)					
Glycaemic Index	-0.493	-0.486	-0.449	-0.454	-0.435					
	(0.006)	(0.009)	(0.019)	(0.020)	(0.030)					
Glycaemic Load	-0.322	-0.219	-0.173	-0.176	-0.219					
	(0.083)	(0.263)	(0.389)	(0.389)	(0.292)					
Carbohydrate (g/day)	-0.236	-0.097	-0.041	-0.039	-0.085					
	(0.210)	(0.622)	(0.841)	(0.851)	(0.687)					
Sugars (g/day)	-0.176	-0.106	-0.118	-0.117	-0.192					
	(0.353)	(0.592)	(0.559)	(0.569)	(0.357)					
Starch (g/day)	-0.137	0.027	0.161	0.181	0.170					
	(0.471)	(0.893)	(0.423)	(0.377)	(0.418)					
Fibre (g/day)	-0.037	-0.025	0.006	0.009	-0.009					
	(0.847)	(0.899)	(0.977)	(0.964)	(0.968)					
Protein (g/day)	0.014	0.124	0.072	0.072	0.093					
	(0.942)	(0.529)	(0.720)	(0.726)	(0.658)					
Fat (g/day)	-0.072	0.172	0.110	0.111	0.199					
	(0.707)	(0.381)	(0.585)	(0.590)	(0.341)					
Energy Intake (KJ)	-0.229 (0.223)	1.000	1.000	1.000	1.000					
Body Mass Index (Kg/m ²)	0.237 (0.207)	1.000	1.000	1.000	1.000					

Table 6.8: Correlation of HOMA_{IR} with dietary glycaemic index, glycaemic load and carbohydrate-related nutrients for South Asian males

HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance; n, sample size

r Spearman correlation coefficient

 r_{c1} , Spearman Partial correlation coefficient; statistically controlled for <u>energy intake (KJ)</u> and body mass index

*r*_{c2},Spearman Partial correlation coefficient; statistically controlled for <u>energy intake (KJ)</u>, body mass index, **age**

*r*_{c3},Spearman Partial correlation coefficient; statistically controlled for <u>energy intake (KJ)</u>, <u>body mass index</u>, age, **physical activity level**

 r_{c4} , Spearman Partial correlation coefficient; statistically controlled for <u>energy intake (KJ)</u>, <u>body mass index</u>, age, physical activity level, **SIMD**

Statistically significant correlation was set at p-value<0.05

		South Asian M	Males (<i>n</i> 30)	
	Glycaemic	Glycaemic	Glycaemic	Glycaemic
	index	load	index	load
	<i>r</i>	r	r _c	<i>r_c</i>
	(<i>p</i> -value)	(p-value)	(p-value)	(<i>p</i> -value)
Glycaemic index	1.000	0.467 (0.009)	1.000	0.652 (0.000)
Glycaemic load	0.467 (0.009)	1.000	0.652 (0.000)	1.000
Carbohydrate (g/day)	0.354	0.972	0.500	0.931
	(0.055)	(0.000)	(0.011)	(0.000)
Sugars (g/day)	-0.005	0.501	0.021	0.286
	(0.980)	(0.005)	(0.919)	(0.165)
Starch (g/day)	0.323	0.678	0.262	0.299
	(0.082)	(0.000)	(0.205)	(0.147)
Fibre (g/day)	0.425	0.536	0.510	0.464
	(0.019)	(0.002)	(0.009)	(0.019)
Protein (g/day)	-0.249	-0.077	-0.254	-0.599
	(0.185)	(0.685)	(0.221)	(0.002)
Fat (g/day)	-0.213	0.246	-0.475	-0.715
	(0.259)	(0.190)	(0.016)	(0.000)
Energy intake (KJ)	0.081 (0.669)	0.735 (0.000)	1.000	1.000

Table 6.9: Correlation of dietary glycaemic index and glycaemic load with carbohydrate related-nutrients and macronutrients for South Asian males (n 30)

 $HOMA_{IR}$, Homeostasis Model Assessment of Insulin Resistance; *n*, sample size

r Spearman correlation coefficient

 r_{c} , Spearman Partial correlation coefficient; statistically controlled for <u>energy intake</u> (KJ), body mass index, age, physical activity level, SIMD

Statistically significant correlation was set at *p*-value<0.05

Table 6.10: Dietary glycaemic index, glycaemic load, carbohydrate-related nutrients and HOMA_{IR} of South Asian males stratified by habitual dietary glycaemic index (lower and upper percentile)

South Asian males (<i>n</i> 30)									
Variable	I	Mean, SD or	Median (IQF	र)	[†] <i>p</i> -value				
	Lower pe GI (ercentile of <i>n</i> 14)	Upper per GI (rcentile of <i>n</i> 16)					
Demographic and anthropon	netric								
Characteristics Age (years)	27.14,	5.27	31.13,	7.52	0.109				
SIMD	2890.00	(1375.25)	4477.00	(782.50)	(0.057)				
BMI (kg/m ²)	23.95	(4.70)	23.25	(3.30)	(0.677)				
Body Fat %	19.96,	7.75	18.06,	4.87	0.423				
Waist circumference (cm)	85.29,	12.13	83.46,	7.90	0.623				
Dietary characteristics									
Glycaemic index	54.00	(2.00)	58.50	(2.75)	(0.000)				
Glycaemic load	144.86,	35.16	166.56,	31.07	0.083				
Carbohydrate (g/day)	266.12,	52.50	289.26,	54.46	0.248				
Sugars (g/day)	80.49	(49.26)	71.54	(29.75)	(0.506)				
Starch (g/day)	162.32,	30.93	197.43,	58.51	0.048				
Fibre (g/day)	14.90	(9.00)	20.43	(12.69)	(0.025)				
Protein (g/day)	86.00,	23.16	82.07,	15.88	0.588				
Fat (g/day)	86.78	(14.27)	82.94	(29.39)	(0.506)				
Carbohydrate (%E)	47.14,	7.22	49.79,	5.31	0.259				
Protein (%E) Fat (%E)	14.85, 36.14,	(5.45) 4.20	14.20 34.34,	(2.18) 4.08	(0.589) 0.245				
Total energy intake (KJ/day)	9033.85,	1292.75	9291.75,	1551.47	0.628				
Biochemical and physiologic characteristics	al								
Fasting glucose (mmol/L)	5.03	(0.61)	4.90	(0.32)	(0.271)				
Fasting insulin (µU/L)	5.38	(3.12)	4.30	(1.50)	(0.032)				
HOMA _{IR}	1.24	(0.70)	0.96	(0.34)	(0.023)				
CRP (mg/L)	0.63	(1.43)	0.65	(1.23)	(0.950)				
¹ Physical Activity Level	1.66,	0.33	1.67,	0.16	0.907				

n, sample size; SD, standard deviation; IQR, interquartile range; SIMD, Scottish Index of Multiple Deprivation rank score; E, energy intake; HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance;

¹calculated from subjects' self-reported physical activity records;

[†]*p*-value not in brackets was calculated by Independent Samples T-Test for normally distributed variables; *p*-value in brackets was calculated by Man-Whitney U test for non-normally distributed variables;

Statistical significance was set at p < 0.05

Verieble	South Asian females (<i>n</i> 22)	European females (<i>n</i> 29)
Variable		
	Prevalence (%)	Prevalence (%)
¹ Highest Education level (university)	81.8	75.9
² British nationality (born in the UK)	50.0	
³ Non-smokers	95.5	86.2
Married	50.0	6.9
Employed	18.2	37.9
Family history of diabetes (answered "yes")	77.3	31.0
Family history of heart problems (answered "yes")	31.8	17.2

¹ for South Asian females; primary school level= 4.5%; high school level= 9.1%; unknown/did not answer=4.5%; for European females; no formal education= 3.4%; high school level= 20.7%

² remaining South Asian females were migrants from Pakistan

 3 for South Asian females; unknown/did not answer= 4.5%

Variable	N	lean, SD or	Median (IQ	!R)	[†] <i>p</i> -value
	South female	South Asian Euro females (<i>n</i> 22) female		opean es (<i>n</i> 29)	
Demographic and anthropom	netric chara	(11.50)	24 00	(11 50)	(0.439)
SIMD	4308.00	(2004 25)	4448.00	(2739 50)	(0.790)
BMI (ka/m ²)	25.25	(9.18)	23.40	(6.15)	(0.106)
Body Fat %	30.65,	9.42	27.55,	8.69	0.230
Waist circumference (cm)	78.74,	12.16	76.26,	10.04	0.429
Dietary characteristics					
¹ Glycaemic index	54.00	(4.25)	54.00	(5.00)	(0.071)
² Glycaemic load	123.00	(23.75)	125.00	(39.00)	(0.797)
Carbohydrate (g/day)	224.98	(40.05)	240.64	(67.34)	(0.506)
Sugars (g/day)	67.94	(31.90)	92.00	(52.93)	(0.074)
Starch (g/day)	152.30,	36.09	123.03,	35.75	0.006
Fibre (g/day)	16.89	(7.42)	16.37	(9.86)	(0.732)
Protein (g/day)	62.80,	11.85	70.41,	18.44	0.080
Fat (g/day)	81.65,	14.15	74.17,	24.85	0.181
Carbohydrate (%E)	48.59,	5.42	48.55,	9.89	0.984
Protein (%E)	13.21,	2.35	14.63,	3.11	0.081
Fat (%E)	38.50,	4.70	34.21,	8.33	0.035
Total energy intake (KJ/day)	7533.83	(810.64)	7296.50	(1243.43)	(0.662)
Biochemical and physiologic	al characte	ristics			
Fasting glucose (mmol/L)	4.70	(0.63)	4.87	(0.96)	(0.962)
Fasting insulin (µU/L)	7.54	4.10	5.45	2.52	0.029
HOMA _{IR}	1.57	0.80	1.16	0.58	0.037
CRP (mg/L)	1.16	(2.64)	0.46	(1.49)	(0.168)
³ Physical Activity Level	1.57	0.12	1.66	0.23	0.052

Table 6.12: Dietary glycaemic index, glycaemic load, carbohydrate-related nutrients and HOMA $_{IR}$ of South Asian and European females

n, sample size; SD, standard deviation; IQR, interquartile range; SIMD, Scottish Index of Multiple Deprivation rank score; E, energy intake; HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance;

¹for SA females; mean GI, SD : 54.59, 2.63 ; EU females; mean GI, SD : 52.79, 3.20

²for SA females: mean GL, SD: 128.68, 26.62; EU females; mean GL, SD : 122.93, 32.70

³calculated from subjects' self-reported physical activity records;

[†]*p*-value not in brackets was calculated by Independent Samples T-Test for normally distributed variables; *p*-value in brackets was calculated by Man-Whitney U test for non-normally distributed variables;

Statistical significance was set at p<0.05

Table 6.13: Sta	aples intake fo	r females	by ethnicity	

Staples intake derived from weighed	Staples intake derived from weighed food records for females (g/day)									
Staples	South A	sian	European females							
	females	(<i>n</i> 22)	(<i>n</i> 29)							
	Mean	SD	Mean	SD						
Unleavened bread	84.71 ^a	80.58	5.35 [⊳]	14.42						
(Chapatti, Naan/Pitta, Paratha)										
Rice (all types)	74.60 ^a	57.12	33.75 ^a	44.41						
Bread	70.37 ^a	38.82	63.85 ^a	49.13						
(White, Wholemeal, Brown)										
Potatoes	51.11 ^a	49.19	51.07 ^a	38.10						
Pasta	50.03 ^a	65.01	79.98 ^a	78.60						
Breakfast Cereals	19.63 ^b	22.14	27.40 ^a	39.76						
(High Fibre, Low Fibre, sugared)										
Bread (Seeded, Rye, Tortilla)	0.00 ^b	0.00	7.80 ^b	18.54						

Different superscript letters indicate significant difference in distributions of staples in the same column (analysed with Friedman's pairwise comparisons test; significance level is 0.05, p< 0.05)

Food Type Amount consumed by females (g/day)								
	Sou	th Asian	(n 22)	Eu	ropean (<i>r</i>	n 29)		
	Median	25 th	75 th	Median	25 th	75 th	<i>p</i> -value	
		PCTL	PCTL		PCTL	PCTL		
Breads								
White bread	64.50	28.52	85.58	28.00	3.22	66.57	0.030	
Wholemeal bread, Brown bread	0.00	0.00	14.45	10.29	0.00	48.27	0.009	
English Muffin, Waffle, Bun (Chelsea)	0.00	0.00	0.00	0.00	0.00	0.00	0.408	
Baguette, Ciabatta, French Stick	0.00	0.00	0.00	0.00	0.00	0.00	0.465	
Seeded bread, Rye bread, Tortilla	0.00	0.00	0.00	0.00	0.00	1.43	0.014	
Pizza	0.00	0.00	44.65	0.00	0.00	28.22	0.746	
Burger, pancake, crumpet, croissant, fruit loaf,	0.00	0.00	9.82	13.00	0.00	41.84	0.055	
granary roll Unleavened bread								
Chapatti (no fat)	42.72	15.00	90.83	0.00	0.00	0.00	0.000	
Chapatti (with fat)	0.00	0.00	0.00	0.00	0.00	0.00	0.251	
Naan/Pitta	0.00	0.00	24.15	0.00	0.00	0.00	0.184	
Paratha	0.00	0.00	3.57	0.00	0.00	0.00	0.008	
Rice								
Rice cooked without oil (Basmati, long grain, brown rice)	37.32	0.00	57.95	0.00	0.00	48.79	0.326	
Rice cooked with oil (Pilau, Biryani)	36.43	0.00	60.12	0.00	0.00	0.00	0.000	

Table 6.14: Amount of carbohydrate foods consumed (g/day) by South Asian (n 22) and European (n 29) females

PCTL, percentile *p*-value for Man-Whitney U test, statistical significance was set at *p*-value<0.05

	Amount consumed by females (g/day)							
Food type	Sou	th Asian	(n 22)	Eu	European (<i>n</i> 29)			
	Median	25 th PCTL	75 th PCTL	Median	25 th PCTL	75 th PCTL	<i>p</i> -value	
Potato								
Potato- fried (chips/fries), boiled, mashed	42.58	0.00	92.71	36.29	9.17	81.05	0.962	
Potato- baked (chips/fries), boiled sweet potato	0.00	0.00	0.00	0.00	0.00	0.00	0.124	
Pasta	37.50	0.00	72.68	54.29	21.52	129.43	0.155	
Noodles	0.00	0.00	0.00	0.00	0.00	0.00	0.541	
Breakfast cereal								
All Bran, Bran flakes, Oatmeal	0.00	0.00	0.00	0.00	0.00	0.00	0.086	
Cocoa Crunch, Frosties, Fruit & Fibre, Shredded Wheat, Special K, Weetabix, Porridge, Muesli	0.00	0.00	14.87	7.86	0.00	34.17	0.144	
Cherrios, Cocoa Pops, Cornflakes, Grapefruits, Puffed Wheat, Ready Brek, Rice Krispies, Rice Pops, Shreddies, Sugar Puffs, Weetos	1.79	0.00	19.32	0.00	0.00	6.50	0.089	

p-value for Man-Whitney U test, statistical significance was set at *p*-value<0.05

	Amount consumed by females (g/day)								
Food type	Sou	th Asian	(n 22)	Eu	ropean (<i>r</i>	n 29)			
	Median	25 th	75 th	Median	25 th	75 th	<i>p</i> -value		
		PCTL	PCTL		PCTL	PCTL			
Cakes	10.29	0.00	37.07	12.50	0.00	24.93	0.831		
Biscuits and crackers									
Biscuits (all types)	7.40	0.00	19.00	10.71	0.00	24.33	0.786		
Digestives, Grain crackers, Oat bicuits, Rich Tea biscuits, Ryvita	0.00	0.00	8.88	0.00	0.00	15.85	0.939		
Cream Crackers, Crispbread Rye, Oatcakes, Cream filled biscuits, Shortbread biscuits	0.00	0.00	5.50	0.00	0.00	10.36	0.353		
Rusks, Wafer biscuits, Pop tarts	0.00	0.00	0.00	0.00	0.00	0.00	0.192		
Snacks									
Snacks (all types)	18.87	0.00	41.04	25.00	9.93	64.04	0.103		
Cereal/fruit/protein bars, Chocolate bars	5.02	0.00	26.95	4.50	0.00	19.07	0.683		
Nuts, trail mix	0.00	0.00	0.00	0.00	0.00	2.71	0.358		
Potato crisps, Corn chips, Raisins, Marshmallow, Popcorn	0.00	0.00	12.05	5.71	0.00	23.72	0.057		
Sweets, fruit gums/Jelly, Jelly beans	0.00	0.00	0.00	0.00	0.00	0.00	0.415		

p-value for Man-Whitney U test, statistical significance was set at *p*-value<0.05

		Amount	consumed	by femal	es (g/day)		
Food type	Sou	th Asian (n 22)	Eu	ropean (<i>n</i> 2	29)	
	Median	25^{th}	75 th	Median	25 th	75^{th}	<i>p</i> -value
		PCTL	PCTL		PCTL	PCTL	
Beverages							
Beverages (all types)	81.38	0.00	233.80	94.29	7.26	321.29	0.695
Lemonade,Oat drinks, Smoothies, Chocolate drink mix, Soy protein drink, Ribena	0.00	0.00	0.00	0.00	0.00	4.40	0.171
Unsweetened fruit juices	0.00	0.00	33.93	0.00	0.00	69.05	0.486
Soft drinks, fruit flavoured drinks, cordials	51.79	0.00	154.03	63.33	0.00	129.79	0.961
Sports drinks	0.00	0.00	0.00	0.00	0.00	0.00	0.101
Dairy (milk, cheese, yogurt)	137.29	44.61	203.89	112.00	42.52	227.29	0.939
Sugar (Sucrose)	5.54	0.00	18.14	0.71	0.00	4.22	0.027*
Fruits	42.86	8.12	83.75	111.29	41.93	195.05	0.013*

p-value for Man-Whitney U test, statistical significance was set at *p*-value<0.05

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Table 6.15: Amount of foods consumed (g/day) with staples by South Asian females (n 22)

			Amoun	t of foods	consume	d with s	aples (g/d	ay)		
Foods eaten with staples	Chapatt Pitta/P	i/Naan/ aratha	Rice		Bread		Potatoes (not including potatoes in curry)		Pasta	
	Median	IQR	Median	IQR	Median	IQR	Median	IQR	Median	IQR
^{1,2} Curry (chicken/lamb/fish)	61.09	109.64	0.00	34.55	0.00	0.00	0.00	0.00	0.00	0.00
^{1,2,3} Curry (lentils, potato)	24.69	56.46	4.50	33.50	0.00	0.00	0.00	0.00	0.00	0.00
³ Meat	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Fish	0.00	0.00	0.00	0.00	0.00	0.00	0.00	18.21	0.00	0.00
Eggs	0.00	0.00	0.00	0.00	0.00	27.77	0.00	0.00	0.00	0.00
² Beans and legumes	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Carrot/ corn/peas/leafy vegetables	0.00	6.43	0.00	1.88	0.00	12.50	0.00	6.16	0.00	3.80
Pumpkin, turnips, swede	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
³ Butter/ cheese/oil	0.00	0.00	0.00	0.00	0.00	5.63	0.00	0.00	0.00	0.00
Jam/honey/syrup	0.00	0.00	0.00	0.00	0.00	3.81	0.00	0.00	0.00	0.00
^{1,2} Yogurt	0.00	7.22	0.00	4.11	0.00	0.00	0.00	0.00	0.00	0.00
Soup	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00

IQR, Interquartile range

Quantity (grams/day) consumed with ¹Chapatti/Naan/pitta/Paratha, ²rice, ³bread (white/wholemeal/brown) ⁴potatoes, ⁵pasta, respectively, were significantly different between South Asian and European females (P<0.05) when tested using Man-Whitney U test

Table 6.16: Amount of foods consumed (g/day) with staples by European females (n 29)

			Amoun	t of foods	consume	d with s	aples (g/d	ay)		
Foods eaten with staples	Chapatti Pitta/Pa	/Naan/ aratha	Rice		Bread		Potatoes (not including potatoes in curry)		Pasta	
	Median	IQR	Median	IQR	Median	IQR	Median	IQR	Median	IQR
Curry (chicken/lamb/fish)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Curry (lentils, potato)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Meat	0.00	0.00	0.00	0.00	4.86	25.21	0.00	31.86	0.00	0.00
Fish	0.00	0.00	0.00	0.00	0.00	11.43	0.00	15.36	0.00	0.00
Eggs	0.00	0.00	0.00	0.00	0.00	18.17	0.00	0.00	0.00	0.00
Beans and legumes	0.00	0.00	0.00	0.00	0.00	11.27	0.00	0.00	0.00	0.00
Carrot/ corn/peas/leafy vegetables	0.00	0.00	0.00	14.97	6.86	27.08	0.00	30.84	0.00	21.47
Pumpkin, turnips, swede	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Butter/ cheese/oil	0.00	0.00	0.00	0.00	10.86	22.54	0.00	6.07	0.00	1.70
Jam/honey/syrup	0.00	0.00	0.00	0.00	0.00	1.77	0.00	0.00	0.00	0.00
Yogurt	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Soup	0.00	0.00	0.00	0.00	0.00	23.72	0.00	0.00	0.00	0.00

	South Asian females (<i>n</i> 22)				
Variables	<i>r</i>	<i>r_{c1}</i>	r _{c2}	<i>r_{c3}</i>	<i>r_{c4}</i>
	(p-value)	(p-value)	(p-value)	(p-value)	(p-value)
Glycaemic Index	-0.069	-0.109	-0.215	-0.211	-0.221
	(0.762)	(0.648)	(0.376)	(0.401)	(0.394)
Glycaemic Load	-0.034	0.166	0.098	0.109	0.096
	(0.882)	(0.485)	(0.691)	(0.667)	(0.713)
Carbohydrate (g/day)	-0.033	0.179	0.260	0.275	0.268
	(0.885)	(0.450)	(0.282)	(0.270)	(0.298)
Sugars (g/day)	0.229	0.457	0.452	0.487	0.486
	(0.306)	(0.043)	(0.052)	(0.040)	(0.048)
Starch (g/day)	-0.138	-0.163	-0.142	-0.137	-0.155
	(0.539)	(0.493)	(0.561)	(0.588)	(0.553)
Fibre (g/day)	0.274	0.270	0.333	0.367	0.465
	(0.217)	(0.249)	(0.163)	(0.134)	(0.060)
Protein (g/day)	-0.113	-0.128	-0.301	-0.301	-0.307
	(0.617)	(0.591)	(0.210)	(0.225)	(0.231)
Fat (g/day)	-0.186	-0.102	-0.378	-0.376	-0.374
	(0.406)	(0.670)	(0.111)	(0.124)	(0.140)
Energy Intake (KJ)	-0.201 (0.370)	1.000	1.000	1.000	1.000
Body Mass Index (Kg/m ²)	0.449 (0.036)	1.000	1.000	1.000	1.000

Table 6.17: Correlation of $HOMA_{IR}$ with dietary glycaemic index, glycaemic load and carbohydrate-related nutrients for South Asian females

HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance; *n*, sample size

r Spearman correlation coefficient

 r_{c1} , Spearman Partial correlation coefficient; statistically controlled for <u>energy intake</u> (KJ) and body mass index

 r_{c2} , Spearman Partial correlation coefficient; statistically controlled for <u>energy intake</u> (KJ), body mass index, **age**

 r_{c3} , Spearman Partial correlation coefficient; statistically controlled for <u>energy intake</u> (KJ), body mass index, age, **SIMD**

 r_{c4} , Spearman Partial correlation coefficient; statistically controlled for <u>energy intake</u> (KJ), body mass index, age, SIMD, physical activity level (PAL)

Statistical significance was set at p<0.05

	South Asian Females (n 22)					
	Glycaemic	Glycaemic	Sugars	Glycaemic	Glycaemic	Sugars
	index	load	intake	index	load	intake
	<i>r</i>	<i>r</i>	<i>r</i>	<i>r_c</i>	<i>r_c</i>	<i>r_c</i>
	(<i>p</i> -value)	(p-value)	(<i>p</i> -value)	(<i>p</i> -value)	(<i>p</i> -value)	(<i>p</i> -value)
Glycaemic index	1.000	0.511 (0.015)	0.139 (0.538)	1.000	0.505 (0.039)	0.026 (0.922)
Glycaemic load	0.511 (0.015)	1.000	0.626 (0.002)	0.505 (0.039)	1.000	0.480 (0.051)
Carbohydrate (g/day)	0.380	0.917	0.641	0.342	0.914	0.509
	(0.081)	(0.000)	(0.001)	(0.179)	(0.000)	(0.037)
Sugars (g/day)	0.139 (0.538)	0.626 (0.002)	1.000	0.026 (0.922)	0.480 (0.051)	1.000
Starch (g/day)	0.462	0.601	-0.100	0.408	0.563	-0.367
	(0.030)	(0.003)	(0.658)	(0.104)	(0.019)	(0.147)
Fibre (g/day)	0.322	0.257	0.205	0.339	0.316	0.041
	(0.143)	(0.248)	(0.360)	(0.184)	(0.216)	(0.875)
Protein (g/day)	0.050	0.151	-0.179	-0.022	0.084	-0.545
	(0.826)	(0.501)	(0.425)	(0.933)	(0.750)	(0.024)
Fat (g/day)	-0.025	-0.102	0.067	-0.257	-0.750	-0.459
	(0.914)	(0.651)	(0.766)	(0.319)	(0.001)	(0.064)
Energy intake (KJ)	0.193 (0.390)	0.492 (0.020)	0.475 (0.026)	1.000	1.000	1.000

Table 6.18: Correlation of dietary glycaemic index and glycaemic load with carbohydrate related-nutrients and macronutrients for South Asian females (n 22)

HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance; *n*, sample size

r Spearman correlation coefficient

*r*_c,Spearman Partial correlation coefficient; statistically controlled for <u>energy intake (KJ), body mass</u> index, age, physical activity level, SIMD

Statistically significant correlation was set at p<0.05

Table 6.19: Dietary glycaemic index, glycaemic load, carbohydrate-related nutrients and $HOMA_{IR}$ of South Asian females stratified by sugar intakes (lower percentile, upper percentile)

		South Asian fe	emales (<i>n</i> 2	2)	
Variable	iable Mean (SD) or Median (IQR)			[†] <i>p</i> -value	
	Lower of sugar	percentile intake (<i>n</i> 11)	Upper p of sugar in	ercentile ntake (<i>n</i> 11)	
Demographic and anthropom Age (years)	etric chara 30.55,	cteristics 7.83	26.55,	5.91	0.191
SIMD	4534.91,	1241.35	4234.18,	1159.47	0.564
BMI (kg/m²)	26.65,	6.19	25.65,	5.54	0.694
Body Fat %	31.42,	9.04	29.87,	10.17	0.710
Waist circumference (cm)	80.55,	13.08	76.92,	11.50	0.497
Dietary characteristics					
Glycaemic index	54.09,	2.77	55.09,	2.51	0.386
Glycaemic load	115.00,	15.15	142.36,	29.10	0.012
Carbohydrate (g/day)	210.37,	23.63	254.83,	48.88	0.013
Sugars (g/day)	53.65	(21.65)	84.50	(56.22)	(0.000)
Starch (g/day)	153.50,	19.49	151.10,	48.50	0.880
Fibre (g/day)	14.96,	3.78	18.84,	7.41	0.138
Protein (g/day)	63.13,	8.29	62.48,	15.03	0.901
Fat (g/day)	81.08,	13.94	82.22,	15.01	0.855
Carbohydrate (%E)	46.35,	4.89	50.83,	5.18	0.050
Protein (%E)	13.87,	(2.24)	12.25	(4.62)	(0.094)
Fat (%E)	39.97,	4.41	37.02,	4.72	0.146
Total energy intake (KJ/day)	7305.80	(631.53)	7822.50	(1435.85)	(0.082)
Biochemical and physiological characteristics					
Fasting glucose (mmol/L)	4.56,	0.37	4.91,	0.39	0.043
Fasting insulin (µU/L)	4.84	(5.30)	7.46	(5.81)	(0.533)
HOMA _{IR}	1.11	(0.99)	1.64	(1.16)	(0.293)
CRP (mg/L)	1.67	(2.82)	0.64	(2.48)	(0.974)
¹ Physical Activity Level	1.56,	0.12	1.57,	0.12	0.901

n, sample size; SD, standard deviation; IQR, interquartile range; SIMD, Scottish Index of Multiple Deprivation rank score; E, energy intake; HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance;

¹calculated from subjects' self-reported physical activity records;

[†]*p*-value not in brackets was calculated by Independent Samples T-Test for normally distributed variables; *p*-value in brackets was calculated by Man-Whitney U test for non-normally distributed variables;

Statistical significance was set at p<0.05

	SFAs (% E)	PUFAs (% E)	MUFAs (% E)
South Asian males	11.50 (8.65,12.85)	6.58, 1.65	10.91, 2.18
European males	11.22 (9.77,14.05)	5.35, 1.63	10.72, 2.13
[†] <i>P</i> -value	(0.460)	0.005	0.745
South Asian females	12.18, 2.90	7.5 (6.09,8.68)	12.58, 2.53
European females	11.13, 3.46	5.53 (4.62,7.16)	11.20, 3.62
[†] <i>P</i> -value	0.259	(0.010)	0.136

Table 6.20: Daily percentage of energy intake from fatty acids by gender and ethnic group. Values shown are Mean, SD or Median (25th PTCL, 75th PTCL))

E, energy intake; SFAs, saturated fatty acids; PUFAs, polyunsaturated fatty acids; MUFAs, monounsaturated fatty acids

[†]*p*-value not in brackets was calculated by Independent Samples T-Test for normally distributed variables; *p*-value in brackets was calculated by Man-Whitney U test for non-normally distributed variables;

Statistical significance was set at P-value<0.05

Table 6.21: Correlation of HOMA_{IR} with dietary saturated fatty acids, polyunsaturated fatty acids and monounsaturated fatty acids in South Asians

	South Asian males and females (<i>n</i> 52) HOMA _{IR}				
Variables	<i>r</i>	r _{c1}	r _{c2}	r _{c3}	r _{c4}
	(p-value)	(p-value)	(p-value)	(p-value)	(p-value)
Total fat	0.018	-0.032	-0.107	-0.096	-0.102
	(0.899)	(0.825)	(0.462)	(0.516)	(0.495)
SFAs (%E)	-0.002	0.003	-0.110	-0.103	-0.116
	(0.989)	(0.983)	(0.452)	(0.488)	(0.438)
PUFAs (%E)	0.119	0.041	-0.068	-0.080	-0.086
	(0.402)	(0.778)	(0.645)	(0.587)	(0.566)
MUFAs (%E)	0.133	0.045	-0.047	-0.032	-0.036
	(0.346)	(0.754)	(0.749)	(0.827)	(0.811)

HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance; *n*, sample size; E, energy intake; SFAs, saturated fatty acids; PUFAs, polyunsaturated fatty acids; MUFAs, monounsaturated fatty acids

r Spearman correlation coefficient

*r*_{c1},Spearman Partial correlation coefficient; statistically controlled for <u>energy intake (KJ) and body</u> <u>mass index</u>

*r*_{c2},Spearman Partial correlation coefficient; statistically controlled for <u>energy intake (KJ)</u>, <u>body</u> <u>mass index</u>, <u>age</u>

 r_{c3} , Spearman Partial correlation coefficient; statistically controlled for <u>energy intake (KJ)</u>, <u>body</u> <u>mass index</u>, age, **SIMD**

*r*_{c4},Spearman Partial correlation coefficient; statistically controlled for <u>energy intake (KJ)</u>, <u>body</u> mass index, age, SIMD, physical activity level (**PAL**)

Statistically significant correlation was set at p<0.05

	South Asian females (<i>n</i> 22) HOMA _{IR}			
Variables	r	r _c		
	<i>(p</i> -value)	<i>(p</i> -value)		
Total fat	-0.107	-0.212		
	(0.635)	(0.415)		
	()	()		
SFAs (%E)	0.295	0.179		
	(0.183)	(0.493)		
	()	()		
PUFAs (%E)	-0.125	-0.510		
	(0.578)	(0.037)		
MUFAs (%E)	0.128	-0.102		
	(0.571)	(0.696)		
Body Mass Index	0.449			
	(0.036)			

Table 6.22: Correlation of HOMA_{IR} with dietary saturated fatty acids, polyunsaturated fatty acids and monounsaturated fatty acids in South Asian females

HOMA_{IR}, Homeostasis Model Assessment of Insulin Resistance; E, energy intake; SFAs, saturated fatty acids; PUFAs, polyunsaturated fatty acids; MUFAs, monounsaturated fatty acids

r Spearman correlation coefficient

 r_{c} , Spearman Partial correlation coefficient; statistically controlled for, total energy intake, body mass index, age, physical activity level and socio-economic status

Statistical significance was set at p<0.05

Chapter 7 General discussion and conclusion

7.1 General discussion

The novel finding of this research is that habitual dietary GI and GL alone did not explain for higher insulin resistance in South Asians compared to Europeans. This is because dietary GI and GL did not differ considerably between South Asians and Europeans. The mean dietary GI of South Asians and Europeans were in the medium GI category (with the exception of European females that had mean dietary GI that was borderline low to medium) and this was similar to the findings of (Goff et al., 2013).

Intakes of some macronutrients, however, differed significantly between ethnicity. South Asian females for instance, had higher intakes of dietary fat compared to European females. In a high fat diet, the dietary GI and GL probably do not have an impact on insulin resistance but rather it is the interaction of dietary fat and other diet variables such as fibre and sugars intake that may affect insulin resistance (Chapter 6). This hypothesis is supported by the observation that in males, dietary GI related negatively to HOMA_{IR}, negatively to fat intakes and positively to fibre intakes. This means the lower the dietary GI, the lower also the fibre intake, the higher the fat intake and the higher the HOMA_{IR}. It is known that fat/oil decrease GI by delaying gastric emptying (Collier et al., 1984) and that fibre lowers glycaemic response (Behall et al., 2006).

South Asian females were found to have higher intakes of dietary fat (Chapter 6) and lower levels of physical activity compared to European females (Chapter 5). In correlation analysis among South Asian females, the higher the carbohydrate (sugars) intakes, the higher the HOMA_{IR} and these relationships were statistically significant (Chapter 6). From this it can be hypothesised that high intakes of total dietary fat (39% of energy from fat), saturated fat (>10% of energy from SFA) and sugars as well as being less physically active all together contribute to insulin resistance in South Asians. Although South Asian subjects' intakes of MUFAs, PUFAs, and SFAs expressed as percent energy (%E) were not markedly

different from that of European subjects, both groups, however, exceeded (%E from SFA in their diet were >10%) recommended population intakes for SFA (Department of Health., 1991). Consumption of dietary SFAs has been shown to positively relate to insulin resistance (Marshall et al., 1997;Manco et al., 2000;Riserus et al., 2007;Isharwal et al., 2009), fasting insulin, and postprandial insulin levels (Parker et al., 1993).

Although SFA intakes did not differ between SA and EU in this current study, South Asians may be more sensitive to the detrimental impact of a high SFA diet compared to Europeans because South Asians are predisposed to insulin resistance and diabetes Type 2 (Bhopal et al., 2014). High SFA diet as opposed to high MUFA diet, have been shown to adversely impact postprandial glycaemic function in subjects predisposed to insulin resistance such as individuals with normal and high fasting triglycerides (Bermudez et al., 2014), in obese and Type 2 diabetics (Christiansen et al., 1997; Summers et al., 2002), obese and insulin resistant (Paniagua et al., 2007) and healthy moderately overweight individuals (Paniagua et al., 2007). Individuals with enhanced risk of type 2 diabetes, daughters of Type 2 diabetic patients for instance, are more sensitive to high intake of SFA in comparison to individuals with lower risks of type 2 diabetes ie. in women with no family history of diabetes (Ntali et al., 2010). Diabetics who were recently diagnosed and individuals with undiagnosed type 2 diabetes have been shown to have significantly higher consumption of saturated fat compared with controls (Thanopoulou et al., 2003; van de Laar et al., 2004).

The study in Chapter 5 confirms findings of previous studies (Fischbacher et al., 2004; Joshi et al., 2007; Ye et al., 2009; Yates et al., 2010; Williams et al., 2011a; Williams et al., 2011b; Ghouri et al., 2013) that physical activity of South Asians is lower than in Europeans and that South Asians and Europeans differ in the types of activities that they habitually perform. Being less physically active may promote insulin resistance because physical activity (Gill and Malkova, 2006; Gill and Cooper, 2008) and cardiorespiratory fitness (Hall et al., 2010; Ghouri et al., 2013) influence insulin resistance, glycaemia and diabetes risk. South Asians especially lacked vigorous activities and structured exercise such as sports which may promote insulin resistance because structured exercise has been shown to improve cardiorespiratory fitness (CRF) (Johannsen et al.,

2013;Giannaki et al., 2015) which in turn improves insulin sensitivity (Larsen et al., 2012a). CRF was identified to be the main factor contributing to insulin resistance in South Asians and they have been shown to be less fit than Europeans (Ghouri et al., 2013).

Furthermore, energy expenditure was found to inversely relate to HOMA_{IR} in South Asians with reported family history of diabetes (Chapter 5) but not in those with no reported family history of diabetes. From this it can be hypothesised that less physically active South Asians who have reported family history of diabetes are more predisposed to insulin resistance (Chapter 5). In line with this, there was an inverse relationship between energy expenditure and HOMA_{IR} in women with family history of diabetes (study subjects were daughters of women with Type 2 diabetes) and this relationship was stronger than for the daughters of women with no family history of diabetes (Higgins et al., 2005).

This study also brings to light the importance of directly measuring the GI of mixed-meals containing significant amount of fats/protein rather than using equations (Wolever et al., 1994) to estimate GI of mixed meals as these equations do not accurately predict the GI of foods (Flint et al., 2004;Dodd et al., 2011). Staples when consumed with curried chicken containing higher amounts of fat and protein were found to illicit lower glycaemic response compared to staples eaten alone (Chapter 6). Foods containing protein and fat have been observed to lower the glycaemic response to the meal (Henry et al., 2006;Moghaddam et al., 2006;Dodd et al., 2011) possibly because fat/oil delay the absorption of food (Collier et al., 1984).

This current study showed that dietary pattern as a whole, along with physical activity is important and may be related to insulin resistance therefore if the GI concept is to be included in dietary guidelines and recommended for healthy people to apply; people need to know what it is, how to make food choices including consideration of GI of foods and how it fits in a healthy diet along with healthy lifestyle. The GI and GL concept may be difficult for the general public to understand and apply in their everyday life. For instance, a "low GI" food does not necessarily mean "healthy" food because low GI foods could also be
high in fat and sugars like ice cream or high in sodium. "High GI" foods in turn, does not necessarily mean that it is "unhealthy" and should be avoided. Watermelon for example, is a high GI food but it is a fruit high in vitamins and antioxidants which is encouraged to consume. More importantly, the quality and quantity of carbohydrates (dietary GL) should be considered. These can complicate existing dietary advice which is probably already challenging as it is for some individuals to understand and apply.

Study strengths and limitations

This thesis included one experimental study which used standard methods to obtain GI values for some staple South Asian foods (Chapter 3). These values were subsequently used in the estimation of habitual dietary GI and GL (Chapter 6) and therefore this deemed dietary GI and GL estimations to be potentially more accurate compared to the use of prediction equations to predict the GI of mixed-meals.

Secondly, the other study was observational in design. To the best of our knowledge, it is the only study focusing on dietary GI, GL and insulin resistance in South Asians in the UK to date. Although it is a small study (n, 111), it nonetheless, was well designed to achieve study objectives using standard methods. There was good evidence that the carbohydrate foods reported by study subjects in the weighed food records (WFR) from which dietary GI and GL were estimated from were habitual (Chapter 4).

A limitation of the observational study on dietary GI, GL and insulin resistance is that it is a relatively small study (n= 111) and recruitment of respondents included non-probability sampling. Therefore the study results may not be representative of the population at large and there may be selection biasness associated with volunteers. Furthermore, underreporting of food and beverage intakes in WFR was evident (68% of possible underreporters) but probably not to a huge extent because all subjects had EI_{rep}:BMR that was equal to or higher than a cut-off of 1.2 which may be a plausible cut-off for very sedentary individuals. When the cut-off was set at lower 99% CL of PAL_{rep}, the EI_{rep}:BMR of

all subjects were well above the cut-off which deemed them to be acceptable reporters of energy intake.

Accuracy of dietary GI and GL estimates are dependent on whether the GI values from International GI tables and other publications that were assigned to the foods in the WFR were correct and most appropriate for the foods. In addition, the nutrient analysis software WinDiets 2005 database did not contain some foods that were reported in the diet of study subjects therefore these were obtained from food packages and other sources hence quality of data is dependent on the correctness of these sources. WinDiets 2005 does not contain particular nutrient values such as dietary fibre and fractions of fat such as saturated fats, polyunsaturated fats and monounsaturated fats for some foods therefore intakes for these nutrients may have been underestimated but probably to a small degree. In recognition of these potential limitations, these results should be interpreted with care and should be confirmed in larger future studies. This study was observational in design and therefore cannot infer causality. Despite these limitations, this small observational study was well designed, applied standard methods, and most importantly, provided interesting hypothesis generating results that contribute to the body of knowledge regarding insulin resistance in South Asians.

Problems encountered in the study

The initial aim of this research was to recruit a representative sample of South Asian subjects in Glasgow for estimation of dietary GI and GL in this population (approximately 152 subjects) but from the early stages it became apparent that recruitment of study subjects was very difficult. There was few response from South Asian subjects to posted study advertisements around Glasgow (public places like shopping centers, shops, university campus) and through email. Consequently a more proactive, personal approach was taken to recruit study participants by speaking to people in public places and handing out fliers of information about the study. Undergraduate students, of which one of them was South Asian in ethnicity, assisted in subject recruitment and provided more links and contacts which encouraged participation in the research. To encourage participation, we also reimbursed subjects' travelling expenses (maximum of 30 pounds sterling per visit) to facilitate visits to the designated laboratory to provide information and blood samples.

Some individuals hesitated or refused to participate in the study because they did not want to give blood samples. Some mentioned that they did not have time to provide blood samples as they were in full-time employment. To overcome this problem, provision of blood samples during subjects' off days from work was arranged for. Some South Asian females expressed reservations in participating due to family commitments (children to take care of and house chores). For these women, the researchers offered them the option of home visits where blood sampling and anthropometric measurements, was arranged at their convenience, to encourage participation. This strategy was effective in some but not all cases.

Ultimately, recruitment by directly approaching subjects and through referrals from study subjects themselves (snowball sampling) was by far, most effective in recruiting South Asian study subjects. Europeans were relatively easier and quicker to recruit, more readily responding to emails and adverts. The necessary measures (home visits) taken did encourage more participation among South Asian females as did assistance from people who had existing links and contacts to potential study subjects as well as reimbursement of travelling expenses.

Another problem encountered during the course of the research was study compliance. There was a relatively high study drop-out rate. In most cases reasons were not given (on ethical grounds, respondents were not obligated to tell the reasons for withdrawing their participation) but in a few cases, women later discovered that they were pregnant and so they no longer fulfilled the study criteria (hormones in pregnancy may influence their fasting glucose and CRP measurements). Two people fell sick during the course of the study rendering them unfit to complete the study. In select cases, study subjects were not contactable for reasons unknown. In light of difficulty in recruitment and to increase study compliance, study requirements for recording of weighed food intakes and physical activity records were made less strict where records were deemed acceptable if completed for at least 3 days as opposed to 7 days as was the initial requirement of the study. This improved study participation and reduced respondent burden which encouraged them to complete the study.

Future studies are required to confirm the findings of this research. Future studies such as this should include the following in the research framework:

- Studies should have a representative sample of the South Asian population in the United Kingdom meaning that the study should include a bigger sample size that is selected randomly in order to obtain more reliable results that can be inferred to the wider population.
- 2. Studies should use a valid dietary assessment method to objectively estimate GI and GL. In epidemiological studies, it is less practical to use weighed food records as data analysis requires an enormous amount of time and effort. However, softwares to analyse dietary GI and GL from weighed food records or the addition of this function to existing nutrient analysis softwares could greatly facilitate data analysis for dietary GI and GL. Such softwares do currently exist but these must be developed to be suitable for the population under study, consider GI of mixed-meals, have clear standard protocol to follow for assigning GI of food items and should be able to analyse data systematically and quickly.
- 3. More objective measures of physical activity (motion sensors device such as accelerometers for instance) should be used to obtain precise measures of physical activity. Such device could also provide the data required to allow researchers to distinguish between light, moderate and vigorous physical activity as well as differentiate between continuous and intermittent activity modes.
- 4. Cardiorespiratory fitness should be accounted for by employing objective measures as there is emerging evidence of the substantial influence of cardiorespiratory fitness on insulin resistance in South Asians.

The findings in this study support current healthy eating and diet advice to avoid excess intakes of dietary fat and to eat a well-balanced diet containing sufficient fruits and vegetables (fibre). These research findings could also provide further important evidence that public health initiatives and interventions should not focus solely on the diet but also give special emphasis on promoting increase of physical activity, particularly vigorous activities like sports and exercise, for the health and well-being of South Asian populations.

7.2 Conclusion

In conclusion, ethnicity (South Asian), the wider diet profile rather than habitual dietary glycaemic index and glycaemic load alone (low GI, low fibre and high fat diets especially SFAs in males for instance; and high fat, high sugar diets in females), low physical activity particularly in structured exercise and sports as well as having a family history of diabetes may contribute to insulin resistance in South Asians. These observations should be confirmed in larger future studies.

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Appendices

- Appendix I : Subject Recruitment advertisements
- Appendix II : Subject Information Sheet
- Appendix III : Consent Form
- Appendix IV : General Questionnaire and Health Screen
- Appendix V : Food Frequency Questionnaire for Carbohydrate Foods
- Appendix VI : Food Inventory (weighed food record)
- Appendix VII : Physical activity record



VOLUNTEERS WANTED for Nutrition Research

We are investigating the Glycaemic Index of selected foods. We need volunteers who are:

- ➤ healthy
- > of any gender
- ➢ aged 20 to 50
- > normal Body Mass Index (please contact us to for an estimate of your BMI)

Study site: Human Nutrition, Division of Developmental Medicine, Yorkhill Hospital, Glasgow, UK, G38SJ

What will the study involve?

- Volunteers will be measured for height, weight, body fat and waist circumference
- Volunteers will be asked to eat selected carbohydrate foods. Fasting blood glucose and changes in blood glucose after foods are eaten will be measured (blood samples from finger prick).

Participate in our study and

.......GET YOUR HEIGHT, WEIGHT, BODY FAT % MEASUREMENTKNOW YOUR IDEAL BODY WEIGHTKNOW YOUR FASTING BLOOD GLUCOSE Travelling expenses will be reimbursed

If you are interested or would like more information, with no obligation to participate,

then please contact:

Mrs. Ramlah George Mohd Rosli at 07518777197 or 01412010502

Email : r.george-mohd-rosli.1@research.gla.ac.uk, Human Nutrition, Division of

Developmental Medicine, Yorkhill Hospital, Glasgow, UK, G38SJ


VOLUNTEERS WANTED for Nutrition Research

We are investigating the habitual diet. Participate in our study and

........FIND OUT ABOUT YOUR DIETGET YOUR HEIGHT, WEIGHT, BODY FAT % MEASUREMENTKNOW YOUR IDEAL BODY WEIGHT

30£ travelling expenses will be reimbursed

We need volunteers who are:

- Of Caucasian origin (Northern European genetic background) or South Asian (of Pakistani ethnicity)
- have lived in the UK for at least 6 months
- > of any gender, height and weight
- > aged 20 to 50

Volunteers will:

- > be briefly interviewed with a food frequency questionnaire/food photo
- > record their habitual food intake and physical activity for up to a week
- > pay **one** visit at Yorkhill Hospital to provide a blood sample and be measured for height, weight and body fat.

If you are interested or would like more information, with no obligation to participate, then please contact

Mrs. Ramlah George Mohd Rosli at 07518777197 or 01412010502

Email : r.george-mohd-rosli.1@research.gla.ac.uk , Human Nutrition, Division of Developmental Medicine, Yorkhill Hospital, Glasgow, UK, G38SJ



SUBJECT INFORMATION SHEET

1. Study title

Glycaemic index measurement of staple foods/meals in the South Asian Diet

2. Invitation

We are pleased to invite you to take part in a research study. Before you decide it is important for you to understand why the research is being done and what it will involve. Please take the time to read the following information carefully and discuss it with others if you wish. Ask us if there is anything that is not clear or if you would like more information. Take time to decide whether or not you wish to take part. Thank you for reading this.

3. Purpose of the study

Given the high level of type 2 diabetes and insulin resistance (which means the body cannot handle the sugar in the blood properly) in South Asian populations and the suggestion that glycaemic index (a way of knowing how fast blood sugar rises after a food or meal is eaten) may play a role in reducing risk of insulin resistance; we would like to determine the glycaemic index of selected staple foods/meals in the South Asian Diet. This study will take 14 days to complete.

4. Why have you been chosen for this study?

We have chosen you because you are healthy (no history of gastrointestinal disorders, diabetes, intake of medication for any chronic disease conditions), not pregnant, not breastfeeding, you do not have intolerance/allergies to any of the test foods or mixed meals, aged between 20 to 50 years and have normal Body Mass Index.

5. Do you have to take part in this study?

It is up to you to decide whether or not to take part in this study. If you do decide to take part you will be given this information sheet to keep and be asked to sign a consent form. If you decide to take part you are still free to withdraw at any time and without giving a reason. If you are a student of the University of Glasgow or any other institution, a decision not to participate will not affect your grades in any way.

6. What will happen to you if you take part?

If you decide to take part in the study, you will be given clear written and verbal instructions on what you need to do. This study will take 14 days to complete (including a gap in-between 7 visits in total). This means that you will have to make 7 visits to the Section of Human Nutrition at Yorkhill Hospitals, Glasgow. Each visit will take two hours. Travelling expenses related to the research will be reimbursed provided they are within reasonable limits. If you consent to participate in this study you will have to do the following:

- 1. You will have to sign the consent form.
- 2. You will be asked to fill in a brief health screen questionnaire.
- 3. Your height and waist circumference will be measured (on the first visit only).
- 4. You will be asked to step onto a machine barefoot which measures your weight, body fat%, and body fat mass (on first visit only).

Before each visit;

You will be asked to come to our lab at Yorkhill Hospital in the morning in a fasting state. This means you will be asked to fast **12 hours** before the visit. For instance, if your visit is at 9.00am, you will be asked not eat or drink anything except plain water after 9.00pm the night before the visit. On the day before your visit, you will also be asked not to consume any alcoholic beverages and caffeine containing drinks and to avoid intense physical activity for long periods of time (e.g. at the gym, excessive swimming, running, aerobics). You will be reminded to follow these instructions through text messages and phone calls.

During each visit;

- 1. When you arrive at the study site at Yorkhill Hospital, the researcher will interview you to confirm if you followed the instructions for the day before.
- 2. You will be told to massage your fingers and warm your hands to ensure good blood flow to your fingers.
- 3. Your finger will be disinfected, left to dry and then pricked to obtain a drop of blood which will be collected on a strip and analysed for blood glucose level. This is your fasting blood glucose sample.
- 4. Then you will be served either a reference food (glucose drink) or one of the test foods/meals (Rice, Pilau Rice, Pilau Rice and chicken curry, chapatti or chapatti and curry) to finish within 15 minutes. This will be served with 200ml of water. Further blood samples will be taken by finger prick at 15, 30, 45, 60, 90, and 120 min after starting to eat. Another 200ml of water will be given during this time. This means that your finger will be pricked for a total of 7 times.

During the 2 hours trial, you will be able to relax and listen to music, read, or bring some work with you.

7. Are there any restrictions during the study?

During the study period, you should not change anything in your diet or your lifestyle. We ask only that you fast for at least 12 hours before your appointment to visit our lab for the glycaemic index measurement of foods trial. On the day before the trial, you will be asked not to consume any alcoholic beverages and caffeine containing drinks and to avoid intense physical activity for long periods of time (e.g. at the gym, excessive swimming, running, aerobics).

8. Possible disadvantages and risks of taking part in the study

Blood sampling during the trials is not expected to cause any adverse effects. You may be concerned about the pain and medical risks associated with drawing of blood. When your finger is pricked for a blood sample, you may feel moderate pain or a stinging sensation. Some people may feel faint and bruising may occur at the site of the needle. We assure you however, that the blood drawing procedure will be conducted by a trained person and that any discomfort for the subject will be minimised by using lancets that are adjustable to the thickness of the skin and the finger prick site will be varied to prevent discomfort at one site. Safety and hygienic measures will be taken at all times during each trial.

9. What are the possible benefits of taking part?

You will receive no direct benefit from taking part in this study. You will however, receive information on your height, weight, body mass index (BMI), body fat percentage and fasting blood glucose.

10. Will my taking part in this study be kept confidential?

All information, which is collected, about you during the course of the research will be kept strictly confidential. You will be identified by an ID number and any information about you will have your name and address removed so that you cannot be recognised from it.

11. What will happen to the results of the research study?

Data from the project will be published only in anonymised form. We aim to write an article on the results and have them published in a scientific journal so that others can see the results and benefit from the information to advance work in the area. You will not be identified in any report/publication.

12. Who is organising and funding the research?

This research is organised and funded by the Department of Human Nutrition, University of Glasgow. The researcher is a student studying for her PhD degree at the Department of Human Nutrition, Glasgow who is sponsored by the Government of Malaysia.

13. Who has reviewed the study?

The project has been reviewed by the Faculty of Medicine Ethics Committee.

14. Contact for Further Information

Should you need further information, please do not hesitate to contact me at my contact details below.

Researcher	:	Ramlah George
Telephone number	:	07518777197 or 01412010502
Email	:	r.george-mohd-rosli.1@research.gla.ac.uk

Or Professor Christine Edwards on 201 0709 c.edwards@clinmed.gla.ac.uk

Thank you very much for taking the time to read this information sheet.



SUBJECT INFORMATION SHEET

1. Study title

Glycaemic Index in the South Asian Diet

2. Invitation

We are pleased to invite you to take part in a research study. Before you decide it is important for you to understand why the research is being done and what it will involve. Please take the time to read the following information carefully and discuss it with others if you wish. Ask us if there is anything that is not clear or if you would like more information. Take time to decide whether or not you wish to take part. Thank you for reading this.

3. Purpose of the study

Given the high level of type 2 diabetes and insulin resistance (which means the body cannot handle the sugar in the blood properly) in South Asian populations and the suggestion that glycaemic index (a way of knowing how fast blood sugar rises after a food or meal is eaten) may play a role in reducing risk of insulin resistance; we would like to determine the glycaemic index of the diet of South Asians in Glasgow. This study will take about 9 days to complete.

4. Why have you been chosen for this study?

We have chosen you because you are representative of either South Asians (from Pakistan) or of Caucasian origin (Northern European genetic background), have lived in Glasgow for at least six months, and is healthy and aged between 20 to 50 years.

5. Do you have to take part in this study?

It is up to you to decide whether or not to take part in this study. If you do decide to take part you will be given this information sheet to keep and be asked to sign a consent form. If you decide to take part you are still free to withdraw at any time and without giving a reason. If you are a student of the University of Glasgow or any other institution, a decision not to participate will not affect your grades in any way.

6. What will happen to you if you take part?

Figure 1 is a flow chart which shows the procedures that you will be involved in on the first visit and Figure 2 is a flow chart which shows the procedures on the second visit.

Visit 1

General Questionnaire

You will be instructed to fill in a general questionnaire (which includes questions on your name, gender, postcode of your address, smoking habit and education background).

Food Frequency Questionnaire (FFQ)

You will be asked to fill in a Food Frequency Questionnaire for staples (bread, cereal and rice) lentils, pulses, dhals and fish and meat.

Food Inventory

We will issue you a 7-day Weighed Food Inventory and let you borrow a food weighing scale to take home with you. The Food Inventory is required to find out what you usually eat. You will be asked to weigh and record all food and drink you take including any leftovers, brand names of products and recipes (ingredients and cooking method) every day for **7 days** (a full week). Written instructions and a demonstration on how to fill in the Food Inventory and how to use the weighing scale will be given.

7 day physical activity record.

We will also issue you with a simplified physical activity record to find out about the kind of physical activities that you do as part of your everyday life. You will use this record sheet to note the time you spend being physically active over 7 days. We will brief you on how to complete the record which you can complete at the same time as recording your 7-day Weighed Food Inventory.

Weight, height and waist circumference measurement

Your weight will be measured using an electronic scale. Your height will be measured using a height measuring scale. For precise height and weight measurements, you will be asked to wear light indoor clothes on the day your measurements are to be made and to take off your shoes before you are measured. Your waist circumference will be measured using a tape measure.

Body fat measurement

Your body fat will be measured using bioelectric impedance analyses (BIA) with a machine called BF-300 TANITA. You will be asked to stand bare feet on the machine. Then a small, harmless amount of electrical current will run through your body. The electricity that enters and exits your body is measured and then used to calculate how much fat you have in your body. This machine is a standard machine similar to those available for purchase by the general public.

Final instructions

You will be asked to make an appointment to return to the Section of Human Nutrition at Yorkhill Hospitals, Glasgow for a second visit. On this second visit, you will be asked to do the following:

- (i) return the weighing scale
- (ii) return the completed Food Inventory
- (iii) return the physical activity record
- (iv) provide a fasting blood sample

Figure 1 : Flow chart of procedures on the first visit (Visit 1)

*Once you arrive at the laboratory, you will take a short rest.

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The area around the puncture site will be cleaned with rubbing alcohol and a wide elastic band will be placed around the upper arm to slightly increase the pressure in the vein.

One end of a sterile double-ended needle that has been attached to an open-ended syringe (which contains an empty test tube) will be inserted into the vein. 5 ml (equivalent to 1 teaspoon) of blood will be collected in an airtight vial or a syringe

The elastic band will be removed and the needle will be withdrawn after blood has been collected. A small cotton ball or pad will be applied with light pressure over the puncture site to stop the bleeding.

After several minutes, the cotton will be discarded or replaced, and a small bandage will be placed on the puncture wound.

End of procedure

Figure 2 : Flow chart of procedures on the second visit (Visit 2)

- Venue : Section of Human Nutrition at Yorkhill Hospitals, Glasgow
- Duration : Approximately 10 minutes in total

*Note: Before your second visit, we will contact you to remind you of your appointment for the second visit to the Section of Human Nutrition at Yorkhill Hospitals, Glasgow.

You will be asked to provide a fasting blood sample on this second visit so this means you will be asked not to eat or drink anything (except plain water) from 9:00PM the previous night until 9.00AM the following morning of your appointment. This is important to make sure your blood glucose measurement is accurate.

7. Are there any restrictions during the study?

During the study, you should not change anything in your diet or your lifestyle. We ask only that you fast for at least 12 hours before your appointment to our lab to have your blood sample taken. It is important that you eat and drink and go about your daily activities as usual during the study.

8. Possible disadvantages and risks of taking part in the study

You may be concerned about the pain and medical risks associated with drawing of blood. When the needle is inserted to draw blood, some people feel moderate pain while others feel only a prick or stinging sensation. Afterward, there may be some throbbing. Some people may feel faint and bruising may occur at the site of the needle. We assure you however, that the blood drawing procedure will be conducted by a trained phlebotomist (a person trained to draw blood) and any discomfort for the subject will be minimised and safety measures will be taken at all times during the procedure.

9. What are the possible benefits of taking part?

You will receive no direct benefit from taking part in this study. You will however, receive information on your height, weight, BMI (body mass index) and body fat percentage.

10. Will my taking part in this study be kept confidential?

All information, which is collected, about you during the course of the research will be kept strictly confidential. You will be identified by an ID number and any information about you will have your name and address removed so that you cannot be recognised from it.

11. What will happen to the results of the research study?

Data from the project will be published only in anonymised form. We aim to write an article on the results and have them published in a scientific journal so that others can see the results and benefit from the information to advance work in the area. You will not be identified in any report/publication.

12. Who is organising and funding the research?

This research is organised and funded by the Department of Human Nutrition, University of Glasgow. The researcher is a student studying for her PhD degree at the Department of Human Nutrition, Glasgow who is sponsored by the Government of Malaysia.

13. Who has reviewed the study?

The project has been reviewed by the Faculty of Medicine Ethics Committee.

14. Contact for Further Information

Should you need further information, please do not hesitate to contact me at my contact details below.

Researcher	:	Ramlah George
Telephone number	:	07518777197 or 01412010502
Email	:	r.george-mohd-rosli.1@research.gla.ac.uk

Or Professor Christine Edwards on 201 0709 c.edwards@clinmed.gla.ac.uk

Thank you very much for taking the time to read this information sheet.

Appendix III



Centre Number: Study Number: Subject Identification Number for this study:

CONSENT FORM

Title of Project: Glycaemic index in the South Asian Diet Name of Researcher: Ramlah George Mohd Rosli

Please initial box

- I confirm that I have read and understand the information sheet dated......
 (version......) for the above study and have had the opportunity to ask questions.
- 2. I understand that my participation is voluntary and that I am free to withdraw at any time, without giving any reason, without my legal rights being affected.
- 3. I agree to take part in the above study.

Name of subject Researcher	Date	Signature						
Researcher	Date	Signature						
Name of person taking consent (if different from researcher)	Date	Signature						

1 for subject; 1 for researcher

Human Nutrition Unit University of Glasgow Appendix IV

Demo	graphic	inform	ation		QUESTIO	NNAIRE			
Name		:	••••••	•••••	••••••	•••••			
Age (y	years)	:	••••••	•••••		•••••			
Gende	er	:	[] male	[]	female				
Marita	al status	:	[] marrie	d	[] single				
Highe	st educa [.]	tion:	[] no forr [] univer	nal sity	education	[] primary	' school	[] high :	school
Home	address	•				••••••			
Telen	hone nur	nher:		•••••		•••••	•••••		•••••
Fmail		•		•••••		••••••			
How	ong have		und in the	l Init	tod Kingdo	·····	· Claso	0.4/2.	
Healt	h Screen	n for St	udy Volun	teeı	rs				
1. At 2. In t *(he	present , the past , ealth prob	do you have y olem suc	i have a *h ou ever ha h as diabet	ealt Id a es, I	th problem health pro heart probl	n? oblem? em, cancer)	yes [] yes []	no [] no []	
3. At 4. Are	present, e you on a (a) at p (b) in t	are yo an advi present he past	u on medio sed diet fo : year	catio or a	on? medical p	roblem,or to	yes [] o lose/ga yes [] yes []	no [] in weigł no [] no []	ht:
5. Ha	ve any of (a) Di (b) He	your ii abetes eart pro	mmediate oblem	fam	ily ever h	ad the follow	wing: yes [] yes []	no [] no []	
6. Do 7. Ha if	you curr ave you e yes , for l	rently s ver smo how lor	moke? oked? ng did you	smo	oke?;	when did y	yes [] yes [] ou stop s	no [] no [] moking?	,
8. Do lf ho	you exe yes , how w long d	rcise? many o you e	times do y xercise ea	ou e ch t	exercise po time?	er week?	yes [] 	no []	

Appendix V

Subject code:

*Food Frequency Questionnaire for Carbohydrate Foods

Please estimate the number of times you have eaten the following foods ON AVERAGE IN THE LAST SIX MONTHS

STAPLES	Medium	Portion	How	ofte	n?		NOTES
	Serving	Size	No	of tim	nes n	(eg brand)	
	Jerving	(Photo		av	ics p	(cg. brand)	
		number	W= 1	uy Nook			
		/	M=M	onth			
		, grams)	N= F	2 2 arely	/Nev	er	
BREADS		514115)	D	W	M	N	
White bread/roll	1 slice	36	-				
Wholemeal bread/roll	1 slice	36					
Chappati/roti with fat	1 chappati	PG13=					
Chappati/roti without fat	1 chappati	PG13=					
Naan/pitta bread	1 naan	133					
Paratha	1 paratha	140					
Puri	1 puri	70					
Baira rotlo	1 rotlo	170					
Papad/papadum	1 nanad	13					
Crisphread- og Rwita	1 crispbread	10					
Dizzo		10					
	T SILCE			\ M /		N	
Low fibro (sugarad	1 bowl	20	U	vv	///	N	
LOW HDIE/Sugared	T DOWL	30					
rice krienice							
	4 h av d	40					
High fibre cereals eg.	I DOWL	40					
Muesti, weetabix	1 have	190		-			
Dhuri (Punjabi oat	I DOWL	180					
porridge)/ Dariya	4	40					
Sweetbreads eg.	I sweetbread	40					
Pancakes, mumn,							
			_	147		N	
RICE	1	D1	U	vv	M	N	
Rice- White, boiled only	1 serv	P1=					
Rice- white, cooked with	1 serv	P1=					
Oll	4	D74					
Vegetable Pilau/Tenri	1 serv	P74=					
Meat Pilau/Ukni	1 serv	P74=					
Meat Biryani	1 serv	P/4=					
Khicheri/Kitchadi (rice &	1 serv	180					
lentils)							
Rice pitta (idli)	1 serv	140					
Dosa	1 serv	90					
Noodles (Chinese)	1 serv	230					
Pasta	1 serv	P2=					
Spaghetti	1 serv	P3=					
Potatoes			D	W	Μ	Ν	
Potatoes (in curry)	1 serv	P45=					
Potatoes (chips/fries)	1 serv	P47=					
Potatoes (baked)	1 serv	P48=					
Pakora	1 pakora						
Others not listed:							

*This Food Frequency Questionnaire was adapted from Kassam-Khamis et al., 1999

LENTILS, PULSES, DHALS	Medium	Portion	How	ofte	n?		NOTES
	Serving	Size	No.	of tin	nes p	er:	(eg. brand)
		(Photo number/gr	D=Da	ay			
		ams)	W= \	Neek			
		(,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	M=M	onth			
			N= R	larely	/Nev	'er	
			D	W	Μ	Ν	
Masoor dhal (red lentil)	1 serv	P70=					
Toovar/Toor dhal (pigeon	1 serv	P70=					
peas)							
Channa dhal (split peas)	1 serv	P70=					
Mung dhal (green lentil)	1 serv	P70=					
Urad dhal (black	1 serv	P70=					
gram)/dhal maash							
Moth dhal	1 serv	P70=					
Whole mung (curry)	1 serv	P70=					
Whole channa (curry)	1 serv	P70=					
(chick peas)							
Khadi	1 serv	P70=					
Dhokra	1 serv	80					
Dhokri	1 serv	80					
Soya beans	1 serv	70					
Kidney beans	1 serv	P51=					
Black eyed beans	1 serv	P51=					
Baked beans	1 serv	135					
Butter beans	1 serv	85					
Guare/cluster beans	1 serv	P65=					
Green/French/runner/br	1 serv	P65=					
oad beans/Uri/Ramay							
Sweetcorn	1 serv	85					
Peas	1 serv	P64=					
Others not listed:							

Instructions for describing portion size (Photo number/grams): Refer to the Food Frequency Questionnaire for Carbohydrate Foods and the photographic atlas for food portion sizes. For each food item that requires portion size description;

- 1. choose the photo which best describes how much you eat per meal or the usual portion size of the food
- 2. WRITE the number/letter ID for the photo you have chosen in the Food Frequency Questionnaire column for 'portion size'

*This Food Frequency Questionnaire was adapted from Kassam-Khamis et al., 1999

FOOD INVENTORY

INSTRUCTIONS

Please record the time, place and meal (breakfast, lunch, dinner, snack) taken every day for 7 days (Monday to Sunday). Weigh and record all food and drink consumed including any leftovers, brand names of products and recipes (ingredients and cooking method) used. For meals eaten away from home, please provide detailed descriptions and approximate amounts of what was eaten. Use household measures (cup, teaspoon, tablespoon) if it is not possible to use the weighing scales.

During the food recording period, it is very important that you eat as you normally would and avoid any temptation to change the diet in order to lose weight or simplify the recording.

Please (i) start a separate page for each day

(ii) start a separate line for each item

Column 1

Record meal (breakfast, lunch, dinner, snack), time, and place of eating

Column 2

Describe each item as accurately as possible, stating where relevant:

- (i) type and brand especially for key staples like rice, chapatti flour, potato
- (ii) whether food is fresh, dried, canned, frozen, salted, smoked, etc.
- (iii) whether food is cooked, if so give method of cooking e.g. fried, baked,
 steamed, stirfried, stirfried and then put to boil until thick etc.. this is very
 important especially for key staples like rice, chapatti flour, potato

Column 3

Record the weight of each item after cooking:

- (i) place scales on a level surface
- (ii) place plate or container on top of scales
- (iii) press 'on/reset' button to turn on scales
- (iv) once zero appears, add first item of food
- (v) record weight displayed
- (vi) press reset button before weighing next item

Wherever possible, record weights in grams. If this is not possible, record weights in household measures (e.g. sugar or jam in teaspoons, stating whether level, rounded, or heaped).

Column 4

Record the weight of any leftovers, such as food remaining on plate, weight of container in which food has been weighed, apple cores, etc.

Columns 5 and 6

Please leave blank.

If you eat a mixed meal/dish rather than a single food, please list each food ingredient and weight on a separate line. For example:

Meat Pilau 100 grams (eaten)

Ingredients(prepared and cooked)basmati rice____ gramslamb breast___ gramsonion___ gramscorn oil___ grams

FOOD INVENTORY - EXAMPLE

Subject code: _____

Date: _____

1. Time/ Place	2. Description of food/drink	3. Weight of food/ Drink (grams)	4. Weight of container / leftovers	5. Please Leave Blank
Breakfast	Toast, white bread (Waburton's)	24		
8.30am	Flora margarine	7		
Home	Coffee (Nescafe)	2		
	Milk, semi-skimmed (Sainsbury's)	10		
	Cornflakes (Kelloggs)	28		
	Milk, skimmed	48		
Snack	Tea	190		
10.00am	sugar	15		
Coffee shop	Scone	48		
Lunch 1.00pm Home	Rice (plain white, basmati)	100		
	beef curry {made with: beef (240g), tomatoes (130g), onions (180g), corn oil (25 g)}	200		
Snack	digestive biscuits	70		
3.30 pm	Tea, strong	140		
Home	milk	50		
	sugar	15		
Dinner	Spaghetti Bolognese - mince beef & dolmio sauce - spaghetti	175 100 75		
	milk	200		
	apple	103		

FOOD INVENTORY - EXAMPLE

Subject code: _____

Date: _____

1. Time/Place	2. Description of food/drink	3. Weight of food/ Drink (grams)	4. Weight of container / leftovers	5. Please Leave Blank
Breakfast	Toast, white bread (Waburton's)	102		
8.30am	Flora margarine	7		
	Mince meat, lamb -stirfried	85		
Home	Tea (Tetly)	1 sachet		
	Milk, semi-skimmed	50		
	Apple	75	9 (core)	
Snack	Tea	190		
10.00am	sugar	15		
Coffee shop	Scone (plain)	48		
Lunch 1.00pm Home	Chapatti (Atta Elephant Medium) - used vegetable oil for wiping	60		
	Potato curry -old potatoes (250g), tomatoes (60g), onions (20 g), cardamom seeds (1g), aniseed (1g), vegetable oil (26g)	200		
Snack	Pakoras (Natco Gram Flour) -fried in corn oil	70		
3.30 pm	Tea (Tetly)	140		
Home	Milk	50		
	Sugar	15		
Dinner	Vegetable pilau -Basmati rice, peas, onions, vegetable oil	250		
Restaurant Mother India's	Lentil dahl -red lentils, large onion, garlic, runner beans, tomatoes, olive oil, cardamom, cloves, cumin	60		
	Milk	200		
	Kheer -milk, sugar, white rice long grain (Sommerfield brand)	30		

FOOD INVENTORY

Subject code: _____

Date:_____

	-	-		_
1. Time/Place	2. Description of food/drink	3. Weight of food/ Drink (grams)	4. Weight of container / leftovers	5. Please Leave Blank

Appendix VII



Physical Activity Record

Subject Id: |__| |__|

to_____

Date: from_____

Developed by: Corinna Koebnick, Jutta Möseneder, Ulrike Trippo, Karen Wagner, Hans-Joachim F. Zunft

German Institute of Human Nutrition (DIFE), Potsdam-Rehbrücke, Dept. Intervention Studies, Nuthetal, Germany, 1st ed., 2003

Dear participants,

In the present study, we need to characterize the lifestyle of all participants as good as possible. Therefore, we ask you to fill in this physical activity record with all your daily activities for a time period of 7 consecutive days. Please, bring the record back after completing.

In order to get information about your habitual lifestyle, it is important that you keep to your usual activities during the recording period. Please report all activities you do accurately and completely.

On the following page you will find instructions on how to fill in this physical activity record. If you need further assistance, please contact us:

Mrs. Ramlah George Mohd Rosli Human Nutrition, University of Glasgow Tel. 0141 201 0486

Dr. Ada Garcia Human Nutrition, University of Glasgow Tel. 0141 201 0570 Thank you for your participation and cooperation!

Instructions

- Please fill in this physical activity record for seven (7) consecutive days

- Common activities and your level of intensity are listed on page 2. To make it simpler the activities are grouped into sleeping time and rest periods, activities at work, leisure time and home activities as well as sports.

- At the top of each day's chart are spaces to fill in for the date, day of the week and the day of recording $(1^{st}, 2^{nd}, 3^{rd}...7^{th} day)$. The day is divided into hours (e.g. 0-1 am, 1-2 am, 2-3 am...11-12pm). Each hour (60 minutes) is split into 4 columns of 15 minutes each.

To start recording activities:

- 1. Turn to the page for the day you are recording (e.g. 1st Day) and fill in the date and day of the week (e.g. Monday, Tuesday etc.)
- 2. Turn to the activity list on page 2 and position it so that it is next to the page for the day you are recording. Make sure the rows of activity and day's chart are perfectly aligned.
- 3. Select the activity you wish to record and move along that row and mark an 'X' in the box of the day's chart at the time you took doing that particular activity.
- 4. If the activity you wish to record is not listed in the activity list provided, try to find an activity on the list similar to it or record it in the 'activities not listed' box.
- 5. It is highly recommended that you fill in the record as soon as you have finished an activity so it is not forgotten or misreported.
- 6. When recording sports activities, please keep in mind, that you should only record the time you are active. If you take a break during sports, record this as 'leisure activity'.

			Da	nte:	_	_	_			Dav o	f the	e we	ek:	_	_	_	_	14	¹ Day
		ACTIVITIES	0	-1 a	m	1 - 2	am	2-3	m	3-4:	m	4	5 am	5-	6 am	6-	7 am	7	-8 am
* Sleeping	time and rest	periods	- Fi	T	ī t	Ť		ΤŤ	T	ŤŤ	T	Ť	TT	ΤŤ	TT	Ť	TT	Ť	TT
* Activiti	es at work		-			-							• •				• •		
Sitting	light work (e.g	desk-work, activities on the computer)																	
	moderate work	(e.g. forklift truck driving, cashier)		+	+	+	\vdash	▋┼┼	+		+	+	++		++	┠┼╴	++	\vdash	++-
Standing	light work (e.g	salesperson in a store, working in a Lab)		-	H	-						-	++-					\vdash	++-
Ŭ	moderate work	c (e.g. filling shelves in a store)			+			╏┼┼			+	+	++		++			\vdash	
	heavy work (e	.g. masonry work)			H								H		++				++-
Walking	slowly	x x /													++			\vdash	
-	briskly								\square				\square		++				
	carrying some	thing (e.g. a tray, dishes)													++				
	carrying heavy	ritems (e.g. boxes, containers)											H		++				
Way to wor	k Watteine	slowly					\vdash						++		++				++
	waiking	briskly							\square				\square		++				
		slowly ¹ (<15 km/h)											++		++		++		
Bic	Bicycling	moderate ² (15 - 20 km/h)			\square				\top				\square		\square		\square		
		fast ³ (20 - 23 km/h)											\square		\square			\square	
		very fast ⁴ (23 - 26 km/h)							\top				\square		\square			\square	
	Driving a ca	ar											\square						
	Travelling i	n a bus, train or car											\square		\square			\square	
Activities n	ot listed:																		
* Leisure	time and hom	e activities																	
Sedentary a	ctivities (e.g. ea	ting, reading, TV viewing, phoning, computer games)																	
Standing ac	tivities (e.g. self	f hair dressing), walking slowly (<4 km/h), shopping																	
Light home	activities (e.g. o	cooking, ironing, dusting), playing a musical instrument																	
Food shopp	ing, child care, (dog walking, fast walking (4-6 km/h),																\square	
Moderate h	ome activities a	nd gardening (e.g. hovering, lawn cutting)																\square	
Home decor	rating and repair	rs (e.g. painting, tiling, wall papering)																\square	
Heavy hom	e activities and	gardening (e.g. digging, shovelling snow)																\square	\square
Very heavy	home activities	and gardening (wood-chopping, carrying heavy logs)																\square	
Activities n	ot listed:																		
* Sports																			
Bowling, pl	aying billiards o	or pool, throwing darts																	
Light gymn	astics, bicycling	- slowly ¹ , horseback riding, table tennis, volleyball																	
Golfing, dar	ncing																		
Aerobics, ba	asketball, bicycl	ing - moderate ² , rambling																\Box	
Badminton,	indoor skating,	rowing, skiing, tennis																	
Hill climbin	ıg, football, han	dball, jogging, bicycling – fast ⁵ , swimming																\Box	
Judo, bicycl	ling – racing ⁴ , so	quash																	
Sports not li	isted:			Τ														Π	