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Dietary intake and factors affecting vitamin D status of Middle Eastern people in the UK

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Dietary intake and factors affecting vitamin D status of Middle Eastern people in the UK

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Dietary intake and factors affecting vitamin D status of Middle Eastern people in the UK

Abstract

Vitamin D is derived through the action of solar ultraviolet B radiation on skin and from a limited number of natural food sources, fortified foods and supplements. It is well known that vitamin D plays an active role for calcium and phosphorus absorption but there is also growing evidence of an association between vitamin D insufficiency and various chronic diseases. Middle Eastern populations are known to be at risk of vitamin D deficiency due to a diet low in vitamin D and low sunshine exposure. Obesity is also a risk factor since vitamin D is sequestered in body fat. This thesis examined dietary intake of vitamin D, obesity and other risk factors for deficiency in Middle Eastern people in the UK.

A questionnaire based survey was undertaken with 242 Middle Eastern respondents. A total of 85% of the sample was estimated to have a vitamin D intake <5 µg/d. Other risk factors for vitamin D insufficiency included covering skin from sunlight (84% females); low use of supplements (18.5%) and being overweight or obese (49% males and 44% females). Vitamin D intake was lowest in those with primary (1.8 µg/d) and secondary school (2.1 µg/d) education compared to higher education (3.6 µg/d). The survey was followed by dietary assessment of 28 Iraqi adults using repeat 24 hour recalls. The results concurred with the survey: mean intake of vitamin D was (3.2±4.4 µg/d) and 78.5% were overweight or obese.

Finally, overweight participants were recruited to observe the effect of fat loss on vitamin D status. Serum 25(OH)D concentrations was measured in Middle Eastern (n=12) and Caucasian adults (n=24). Firstly seasonal changes were

observed between October and January (with no weight loss). Then participants were advised on weight reduction to observe the effect of fat loss on serum 25(OH)D. Vitamin D deficiency (<25 nmol/l) was observed in 67% of the Middle Eastern group in October increasing to 92% in January. Of the 36 participants, only 17 lost ≥1kg of fat mass between January and April. No difference was found in serum 25(OH)D between those that lost fat mass and those that did not, and no correlation was found between the amount of fat lost and change in 25(OH)D. In the total sample, there was a negative association between serum 25(OH)D and waist circumference and waist-hip ratio, but no correlation was found between 25(OH)D and fat mass, thus indicating a relationship with visceral fat stores rather than total fat mass.

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Dedication

This thesis is dedicated to my late father and my late grandfather

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Author's declaration

At no time during the registration for the degree of Doctor of Philosophy has the author been registered for any other University award without prior agreement of the Graduate Committee. This study was financed with the aid of Ministry of the Higher Education and scientific research/Iraq.

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- Ahmed, W. A., Rees, G. A., de Looy, A. (2012). Seasonal variation in vitamin D status in overweight and obese people of Iraqi and Caucasian descent.
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CHAPTER 1: Introduction

1.1 Background

Vitamin D deficiency is common in Middle Eastern regions despite abundant sunshine (12- 42°N) (El-Hajj Fuleihan 2009). Additionally, it has been found that immigrants from the Middle East to other countries world-wide are at risk of vitamin D deficiency (Glerup et al. 2000, Holvik et al. 2004, Erkal et al. 2006, Hobbs et al. 2009, Madar et al. 2009, Brock et al. 2010a, Pinelli et al. 2010). Limited data are available from the UK about vitamin D status among Middle Eastern people and about risk factors such as: dietary intake, lifestyle, and obesity in this group. Thus, this thesis will examine risk factors for vitamin D deficiency in Middle Eastern people using a self-completion survey and repeat 24 hour dietary recalls. As overweight and obesity were found to be common in the participants of these preliminary investigations, an experimental phase is then presented which examined the relationship between body fatness and vitamin D status, and the effect of weight reduction on vitamin D status. The rest of this chapter introduces the aims and objectives of each chapter in this thesis.

Chapter 2: A review of literature

This chapter is a review of the literature concerned with vitamin D. The first three sections include a definition of vitamin D, vitamin D synthesis and sources, and current recommendations for vitamin D intake. Metabolism and functions of vitamin D are summarized, and links between vitamin D and chronic diseases including obesity are briefly discussed. The determination of vitamin D status from plasma is reviewed and the effect of seasonal variation and extent of hypovitominosis is discussed in different groups. Finally, vitamin D toxicity and an overall conclusion are summarized.

Chapter 3: Vitamin D intake and other risk factors for vitamin D insufficiency in Middle Eastern people living in the UK: A comparison of cultural and ethnic groups

In this chapter a “Food and Health Questionnaire” was designed to obtain information about factors that can affect vitamin D status such as eating habits, sunlight exposure, Body Mass Index (BMI) and demographic characteristics. Participants included Middle Eastern students, refugees and immigrants living in the United Kingdom. Questionnaires were administered via interview or posted to the participants. Two hundred and forty two volunteers (n=167 males) were included.

3.1 Aims

The aim of this chapter is to compare factors affecting vitamin D status and estimated dietary intake of vitamin D between different cultural and ethnic groups from the Middle East now living in the UK.

3.2 Objectives

The objectives of this chapter are to:

- Assess dietary intake of vitamin D, and detect any possible lower intakes in the sample.
- Explore the relationship between lifestyle and vitamin D intake.
- Establish the prevalence of overweight and obesity.

Chapter 4: Dietary intake of vitamin D and calcium in Middle Eastern people living in the UK

In this chapter twenty eight Iraqi adults (n=16 males) were recruited from University of Plymouth. Dietary intake was estimated 3 times using a 24 hour dietary recall. Portion size was estimated using household measures and the Photographic Atlas of Food Portion Sizes (Nelson et al. 1997a). Energy and macronutrients estimations were calculated using CompEat nutritional analysis programme. Weight and height were measured to calculate Body Mass Index (BMI). Under-reporting of energy intake was calculated and compared to estimated BMR using the Goldberg equation.

4.1 Aims

The aim of chapter 4 is to obtain an estimate of energy and macronutrient intake of Middle Eastern people living in the UK

4.2 Objectives

The objectives of chapter 4 are to:

- Obtain a record of all food/drink participants consumed within proceeding 24 hours
- Assess dietary intake of calcium and vitamin D
- Calculate BMI and relate to nutrient intake

Chapter 5: The relation between body fatness and vitamin D status of Middle Eastern people and Caucasians living in the South West of the UK

This chapter is an experimental study to further investigate the relationship between vitamin D status and body fat. Twelve Middle Eastern (n=6 males) and 21 Caucasian (n=3 males) volunteers were recruited from University of Plymouth. The entire study sample had a high percentage of body fat; >20% for males and >30% for females. The effect of seasonal variation of serum vitamin D was measured over 3 months (October-January) and then participants were advised on weight reduction for 3 months (January-March/April).

5.1 Aims

The aims of this chapter are to:

- Examine the effect of seasonal variation over a 3 month period of reduced daily sunlight (October-January) on serum 25(OH)D concentrations in Middle Eastern people in south west of the UK compared to Caucasian group.
- Present the relationship between vitamin D status and body fatness
- Assess the effect of moderate loss of body fat induced by diet and increased physical activity on serum 25(OH)D concentrations and insulin levels.

5.2 Objectives

The objectives of chapter 5 are to:

- Assess vitamin D status [serum 25(OH)D concentrations] and compare it between Middle Eastern and Caucasian subjects.
- Investigate seasonal variation on vitamin D status in Middle Eastern and Caucasian subjects.
- Explore the relation between body fatness and vitamin D status.
- Assess the effect of fat loss by diet and exercise on vitamin D status and insulin levels in Middle Eastern and Caucasian participants.

Chapter 6: Dietary sources of vitamin D and dietary assessment methods

In this chapter the most important dietary sources of vitamin D were identified from the food records of Middle Eastern subjects (n=28 and n=12) in chapters 4 and 5, respectively. The validity of the food questionnaire (chapter 3) for estimating vitamin D intake in Middle East adults was assessed by comparison to the use of three 24 hour recalls (chapter 4) and a 3 day diet diary (chapter 5).

6.1 Aims

The aims of this chapter are to:

- Identify the most important dietary sources of vitamin D assessed using three 24 hour dietary recalls (chapter 4) and using a 3 day food diary (chapter 5).
- Evaluate the FFQ with 31 items for rapid assessment of vitamin D intake in Middle Eastern adults living in the UK (used in chapter3).

6.2 Objectives

The objectives of chapter 6 are to:

- Collect detailed information from 24 hour dietary recalls (analysed by CompEat) in chapter 4 and food diary (analysed by Dietplan6) in chapter 5 on foods consumed by Middle Eastern subjects to indicate the most important dietary sources of vitamin D.

- Compare the assessment of vitamin D intake by using the FFQ to assessment by 24 hour dietary recall and food diary.

CHAPTER 2: Literature Review

2.1 Vitamin D

Vitamin D (calciferol) is a fat soluble nutrient. It is found in natural dietary sources, fortified foods and supplements and it can be obtained from the action of sunlight on the skin. Vitamin D is not strictly a vitamin; it is technically a hormone when produced by the body and it is technically a vitamin when supplied by the diet (Grodner et al. 2012). Regardless of this, it is an essential substance that plays an active role in maintaining good health primarily for calcium and phosphorus absorption (Barasi 1997). There are two chemical forms of vitamin D; vitamin D_2 (Ergocalciferol), which is synthesized when ergosterol (provitamin) in plants is irradiated by UV light (Lutz and Przytulski, 2001) and vitamin D_3 (Cholecalciferol), which is formed through the action of UV light on 7-dehydrocholesterol (provitamin) in the skin (Holick 2005). Vitamin D_3 is the form most commonly found in natural dietary sources, in contrast to vitamin D_2 (Brody 1999). Both forms of vitamin D have equal effectiveness for humans and are used for food fortification and for supplements (Holick 2005, WHO and FAO 2006).

2.2 Metabolism and functions of vitamin D

Once vitamin D (either D_2 or D_3) is absorbed from the diet in the gut or made in the skin, the vitamin is stored in the body fat (Bates et al. 1997) or it enters the circulation and is bound to the group-specific protein, known as vitamin D-binding protein (DBP), for transport in the blood to the liver (Holick 2005). The liver converts the vitamin to 25-hydroxyvitamin D [25(OH)D], also called calcidiol by enzyme action and then the kidney alters 25(OH)D to 1,25-dihydroxyvitamin D [1,25$(OH)_2$ D] (calcitriol) which is the biologically active form of vitamin D (DeLuca 2004) . This production of 1,25 $(OH)_2$ D is controlled via

parathyroid hormone (PTH) (Figure 2.1). Vitamin D plays a major role for calcium and phosphorus absorption, bone growth, muscle function (DeLuca 2004).

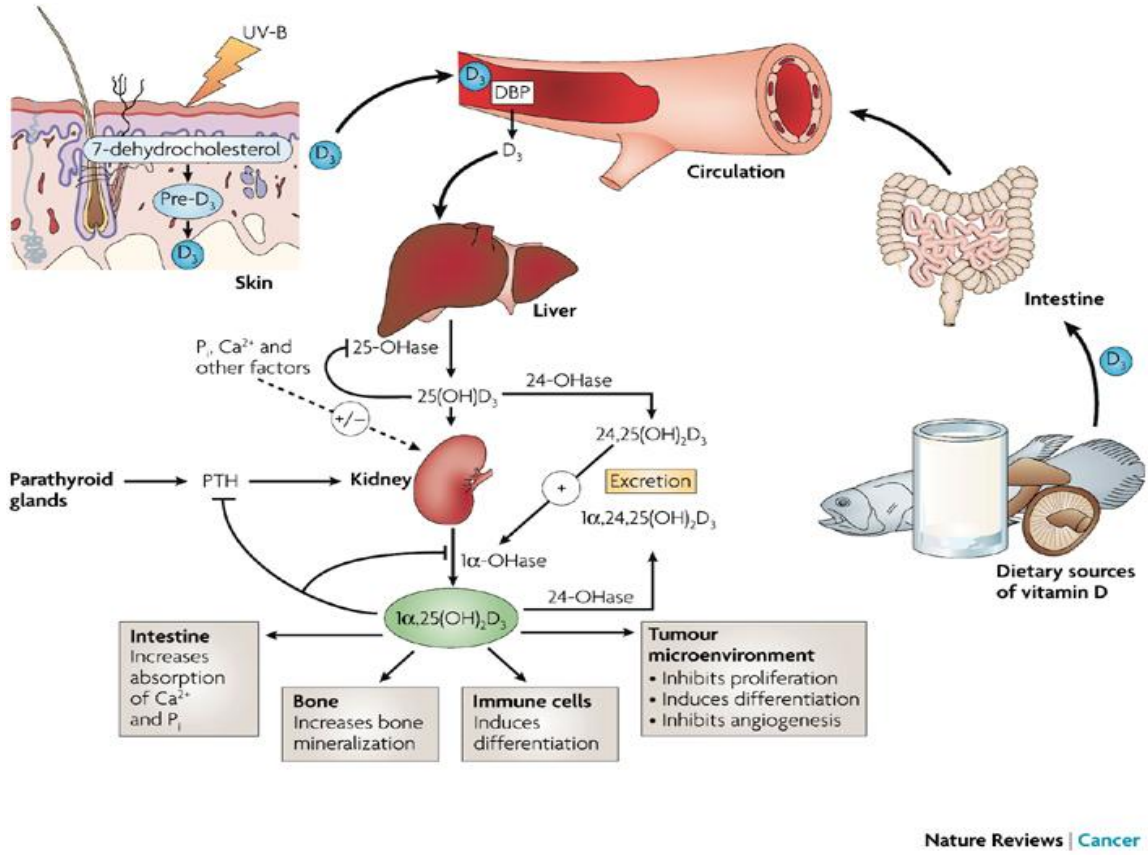

Figure 2.1 Vitamin D metabolism (Deeb et al. 2007)

2.3 Vitamin D synthesis and sources

2.3.1 Sunlight

During exposure to sunlight, vitamin D_3 is synthesised through the action of solar ultraviolet B photons [UVB radiation (290-315 nanometres in wavelength)] on 7-dehydrocholesterol in the skin leading to its transformation to pre vitamin D_3, which is rapidly converted to vitamin D_3 (Holick 2011). Whereas, plants exposed to UV light convert ergosterol (provitamin) to vitamin D_2 (Brody 1999).

Most humans obtain most of their vitamin D from casual exposure to sunlight (Holick 1994). Several factors influence the cutaneous production of vitamin D, these include: geographic latitude, season (which effects cloud cover), time of day, smog (air pollution), sunscreen use, skin colour (darker skin pigmentation reduces the effect of UV on the skin), mobility and aging (ability of older adults or people with disabilities to get outdoors and aging may decrease the amount of vitamin D formed in skin from sunlight exposure) and cultural clothing customs that hide the body (Bates et al. 1997, Grodner et al. 2012).

2.3.2 Natural dietary sources

Vitamin D is not found in plant materials (e.g. vegetables, fruits, or grains) (DeLuca 2004) and it is present naturally in few sources of animal-related foods such as: egg yolks, liver, oily fish (salmon, mackerel, herring and sardines) and cod liver oil (Holick 2004, Grodner et al. 2012). The vitamin D content in fish varies widely even within species. Interestingly, research has shown that farmed salmon, contained about one quarter of the vitamin D found in wild salmon (Lu et al. 2007).

2.3.3 Fortified foods

When sunlight exposure is scarce, oral intake of vitamin D, either dietary or supplementary becomes essential in order to maintain sufficient 25(OH)D.

However, since natural dietary sources of vitamin D are limited, fortification with vitamin D of certain food stuffs has been accepted as a strategy to improve serum 25(OH)D concentrations. Milk and other dairy products are a good vehicle for vitamin D fortification, because they contain calcium and phosphorus, which need vitamin D for absorption (Grodner et al. 2012) . On the other hand, many individuals are vegetarians and others may have lactose intolerance, therefore additional foods may need to be fortified with vitamin D such as: breakfast cereals, orange juice (Tangpricha et al. 2003) and bread (Natri et al. 2006).

In the UK, margarine, some breakfast cereals, some processed milk and some powdered milk are fortified with vitamin D (Food Standards Agency 2003), whereas in the US almost all fluid milks, 75% of ready-to-eat breakfast cereals, slightly more than half of all milk substitutes, approximately one-fourth of yogurts, and 8–14% of cheeses, juices, and spreads are fortified with vitamin D (Yetley 2008).

2.3.4 Supplements

Vitamin D is present in a range of licensed medicines and food supplements (including fish oil products and calcium supplements), at levels up to 12.5 µg dose (Food Standards Agency 2003). Moreover, there are a variety of multivitamins that contain 10 µg which are available as capsules and tablets (Holick 2005) or as an oil preparation that contains 100.000 IU/ml (Holick 2006). All forms of supplements are thought to provide the same levels of bioavailability (Dowd and Stafford 2008).

2.4 Recommendations for vitamin D intake

2.4.1 Dietary recommendations for vitamin D

Vitamin D was originally measured in International Units (IU), which is the older quantitative unit of biological activity for vitamin D before the pure compound was isolated; an IU is equal to 0.025 µg of cholecalciferol (Mann and Truswell 1998).

A daily intake of 5µg of vitamin D is recommended by FAO and WHO (2002) for healthy adults aged 50 y and younger, while 10 and 15 µg were recommended for the age groups 50-65 and >65 y, respectively for those without predominant exposure to sunlight. The UK has no dietary recommendation for vitamin D; unless pregnant, lactating or have no sunlight exposure and then 10 µ/d recommended (Department of Health 1991).

2.4.2 Recommended sunlight exposure

Various durations of exposure to sunlight have been recommended because of the differences in the degree of sunlight in various countries and the individual and racial variances in production of vitamin D. It has been suggested that light skinned adults can obtain 5 µg of vitamin D by exposing their hands, arms, and face to sunlight for 15 minutes twice a week (Lutz, 2001 .354), whereas the elderly should have 30 minutes of daily sunlight on the face and legs at a latitude of 37°N. However, 1-2 h may be necessary in the north of Britain (Eastwood 2003). Dark skinned adults require 5-10 times the exposure that a white person requires to synthesize the same amount of vitamin D (Holick 2011).

2.5 Vitamin D linked to chronic diseases

Recent studies have revealed that vitamin D has a role in: regulating the immune system, regulating the blood pressure, protects against cardiovascular heart disease, and controls cancer cell growth (Figure 2.2).

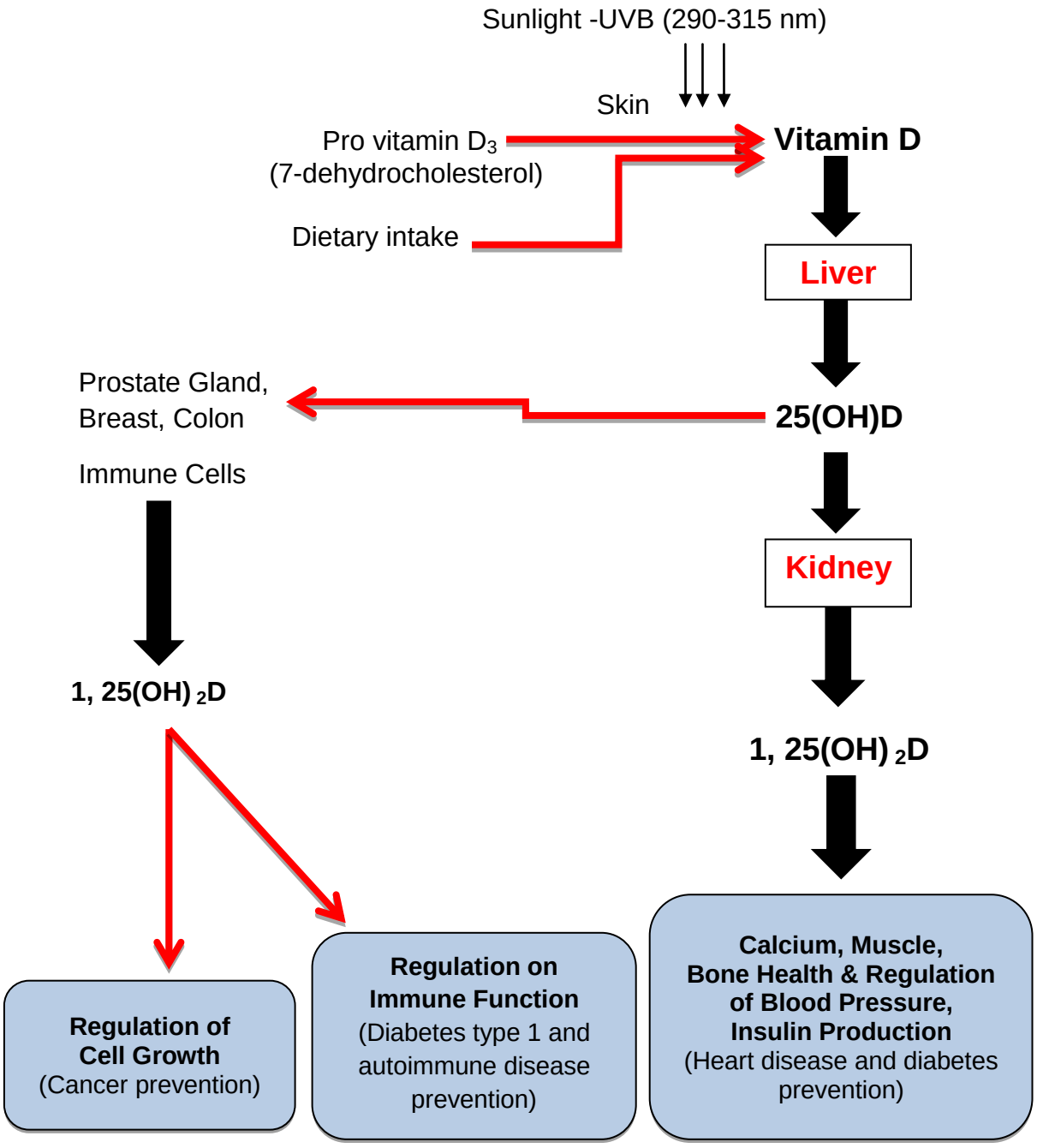

Figure 2.2 Schematic representation of the metabolism and the multitude of other potential physiological actions of vitamin D. Adapted from (Holick 2006)

2.5.1 Body fat and obesity

There is an inverse relationship between serum 25(OH)D concentrations with fat mass, and percentage of body fat (Valiña-Tóth et al. 2010) and BMI (Buffington et al. 1993, Wortsman et al. 2000, Arunabh et al. 2003, Snijder et al. 2005, Bischof et al. 2006, Carlin et al. 2006, Truesdell et al. 2011) (See chapter 5).

Since serum 25(OH)D is negatively related to body fat and BMI, it has been suggested that BMI should be taken into consideration when determining vitamin D requirements; this is due to excessive storage of vitamin D in the adipose tissue.

Bischof and others (2006) found that, serum 25(OH)D concentrations negatively correlated to BMI in 483 subjects (age 48.2±16.0 y). Additionally, the prevalence of vitamin D deficiency (serum 25(OH)D concentrations <22.0 nmol/l) increased from 8.8% in subjects with BMI<30 kg/m^2 to 15% in subjects with BMI>30 kg/m^2.

Another research group (Lee et al. 2009b) indicated that, efficacy of vitamin D supplementation is dependent on BMI and the overweight and obese subjects with hypovitaminosis D might require higher doses of vitamin D to achieve vitamin D repletion compared with individuals with normal body weight; in a study that recruited 95 subjects with vitamin D deficiency (serum 25(OH)D concentrations <15 nmol/l). The subjects received 250 µg/d of oral vitamin D supplement (cholecalciferol) for 1 week. The results indicated that, serum 25(OH)D concentrations correlated negatively with BMI. Moreover, Jorde and others (2010) included 93 adults in an intervention study; the subjects received 1000 µg/week vitamin D supplement (cholecalciferol) for 12 months. The results

indicated that, serum 25(OH)D concentrations correlated negatively with BMI both at baseline and after 12 months.

Forsythe and other researchers conducted two studies about the response of vitamin D supplements and body composition (weight, BMI, waist circumferences, fat mass and fat free mass, etc.) in healthy adults. In the first study (2009), a total of 237 volunteers (age 20-40 y) were randomised to receive 5, 10 or 15 µg cholecalciferol or placebo daily for 22 weeks during the winter months (October-March); fat mass was significantly associated with the serum 25(OH)D concentrations response to supplementation in males. However, no such association was found in females. In the second study (2012), a total of 110 young Irish adults (age 20-40 y) and 102 older Irish adults (age ≥64 y) received a daily dose of 15 µg cholecalciferol or placebo during October. The results indicated that, BMI associated negatively with the change in serum 25(OH)D concentrations in older adults. However, no such associations were found in younger adults.

2.5.2 Immune system

- **Metabolic syndrome and diabetes mellitus**

Many studies have shown an inverse association between 25(OH)D and the metabolic syndrome (Ford et al. 2005, Botella-Carretero et al. 2007, Hyppönen et al. 2008, Lee et al. 2009a, Pinelli et al. 2010, Kayaniyil et al. 2011).

A total of 83,779 healthy female nurses (age 30–55 y), living in 11 US states have recruited in a large prospective study. The results suggested a potential beneficial role for vitamin D intake in reducing the risk of type 2 diabetes mellitus (Pittas et al. 2006). Also, Knekt and others (2008) demonstrated that, the high serum 25(OH)D concentrations provide protection against type 2 diabetes.

Additionally, Mattila and colleagues (2007) found a significant inverse relationship between serum 25(OH)D concentrations and the risk of type 2 diabetes mellitus in 4097 adults (age 40-69 y) in Finland. While McGill and other researchers (2008) found an inverse association between serum 25(OH)D concentrations and markers of type1 diabetes mellitus in 250 adults (age >18 y) with BMI 28–50 kg/m^2 in New Zealand. Moreover, it has been found that a low serum 25(OH)D concentrations is associated with a high risk of diabetes mellitus and the concentration is inversely associated with insulin resistance in overweight or obese 5787 Korean adults (age >20 y) (Choi et al. 2011).

- **Other autoimmune diseases**

It has been found that high serum 25(OH)D concentrations are associated with a lower risk of multiple sclerosis and has a protective effect on risk of developing the disease (Munger et al. 2004, Soilu-Hanninen et al. 2008). In addition, another study indicated a high prevalence of vitamin D deficiency in multiple sclerosis patients (Yildiza et al. 2011). Also new studies established a link between low serum 25(OH)D concentrations and human immunodeficiency virus with the risk of severe disease, acquired immune deficiency syndrome, and high mortality (Dao et al. 2011, Giusti et al. 2011, Vescini et al. 2011, Viard et al. 2011).

2.5.3 Hypertension and cardiovascular disease

A scientific link between low serum 25(OH)D concentrations and incidence of hypertension has been established following several recent studies confirming the relationship (Forman et al. 2007, Barnard and Colón-Emeric 2010, Al Mheid et al. 2011, Burgaz et al. 2011).

Intervention trials indicate that vitamin D may protect against risk of heart diseases, as it has been found that low 25(OH)D is associated with incident

cardiovascular disease (Giovannucci et al. 2008, Wang et al. 2008, Kilkkinen et al. 2009, Anderson et al. 2010, Barnard and Colón-Emeric 2010).

The role of sufficient vitamin D in prevention of hypertension and cardiovascular heart disease is that 1,25$(OH)_2$D is one of the most potent hormones to down regulate the blood pressure hormone renin in the kidney. Though, the exact mechanism of this role is not fully understood. (Li et al. 2002).

2.5.4 Cancer

There is strong evidence indicating a relationship linking vitamin D Deficiency to risk of some cancers. It has been found that vitamin D intake has a protective effect on risk of developing colorectal cancer (La Vecchia et al. 1997, Gorham et al. 2005), and another study indicated that low 25(OH)D may increase risk of breast cancer in a UK Caucasian population (Lowe et al. 2005).

In 2006 a study conducted by Skinner and others pointed that higher intakes of vitamin D were associated with lower risks for pancreatic cancer. In addition, Li and others (2007) indicated that suboptimal 25(OH)D may play an important role in preventing prostate cancer progression. The explanation of this is that 25(OH)D is used by the prostate cells to make 1,25$(OH)_2$D, which helps regulate prostate cell proliferation and thus decreases the risk of prostate cells to becoming malignant (Feldman et al. 2000, Chen and Holick 2003, Holick 2004b).

A new study by Thomas and others (2011) found that lower 25(OH)D appeared related to a progressive stage of hematological disease (leukemia) and poor response to therapy. Moreover, Lappe and other researchers (2007) demonstrated that improving 25(OH)D substantially reduces all-cancer risk in postmenopausal women.

The role of vitamin D in cancer prevention has been explained by Holick (2005); VDR exists in most tissues in the body and 1,25$(OH)_2$D is a potent inhibitor of both normal and cancer cell growth (Feldman et al. 2000, Chen and Holick 2003, Holick 2003, 2004b). It is proposed that the increased renal production of 1,25$(OH)_2$D could in some way regulate cancer cell growth and therefore mitigate the cancer's activity.

2.6 Determination of vitamin D status

2.6.1 Evaluation of dietary intake of vitamin D

Dietary intake assessment is part of the process of determining nutritional status. The assessment may reveal the likelihood of developing nutrient deficiencies (caused by an adequate intake of a nutrient) or excesses. Nutritional assessment evaluates the dietary intake of the foods each person eats to determine the quantities of nutrients consumed as compared with the recommended (Grodner et al. 2012). Methods for measuring food intake may include:

- **Description weighted and estimated records:** in this method, subjects can be taught to describe and give an estimate of food weighed before eating and then to record any leftover.
- **Diet histories:** in this method, subjects are asked to remember accurately the frequency and quantities of food eaten at a previous time. The diet history, like the questionnaire, is a repeatable and relatively valid method. It covers significant periods and so compensates for the possible misrepresentations due to week-to-week variations in diet. This is particular importance for vitamin D. A diet history consists of:
- Cross-check food frequency list
- 24 h dietary recall

- 3-day food diary
- Detailed interview to measure amounts and frequency of a wide variety of foods (Eastwood 2003).

After the collection of food consumption data, it can be analysed through several computer dietary analysis software packages to convert food to nutrient intake and compare this with the recommended. When this analysis is performed on a sample of subjects representative of the bigger population, the estimation of nutrient intake can be the indicator of the nutritional status of the population (Grodner et al. 2012).

2.6.2 Assessment of vitamin D status

A deficiency is accurately diagnosed by measuring the concentration of a specific form of vitamin D, which is serum 25(OH)D concentration as the best indicator or barometer of vitamin D status in blood (DeLuca 2004).

The deficiency cut-off for vitamin D status (serum 25(OH)D concentrations) is set to be <25 nmol/l as a sufficient concentration to prevent the severe hypovitaminosis D which leads to soft bone tissue (rickets in children and osteomalacia in adults), but not in relation to other health outcomes (Mavroeidi et al. 2010). Additionally, even less severe forms of hypovitaminosis D have short- and long-term health implications, and therefore, concentrations of ≥75 nmol/l were identified as necessary for optimum bone health (Dawson-Hughes et al. 2005). Table 2.1 below shows vitamin D related diseases and serum 25(OH)D concentrations.

This cut-off was based on a threshold required for a range of functional outcomes, including maximal suppression of circulating parathyroid hormone,

greatest calcium absorption, and highest bone mineral density (Hyppönen and Power 2007).

Table 2.1 Vitamin D-related bone disease and 25(OH)D

Bone disease	**25(OH)D** (nmol/l)
Rickets / Osteomalacia	0-25
Osteoporosis	25-75
Normal	75-150

Adopted from (Heaney 2004)

2.7 Vitamin D and seasonal variation

Solar radiation is weaker and hours of sunlight are shorter in winter compared with summer and in high latitudes compared with low latitudes (Levis et al. 2005). Thus, seasonal variation has a major effect on 25(OH)D among individuals living in Europe compared with regions closer to the equator and there is a significant seasonal variation, with a higher 25(OH)D concentrations at the end of summer and lower concentrations at the end of winter (Tjellesen and Christiansen 1983, Vanderschueren et al. 1991, Aguado et al. 2000). In contrast, people living in sunny regions are also at risk of low 25(OH)D, as a result of covering up with clothing and the increased use of sunscreens and the filtering of UV waves in automobile glass, because of the widespread sensitivity to skin cancer and sun exposure (Horani et al. 2011).

2.8 Vitamin D deficiency and insufficiency

Vitamin D deficiency occurs among individuals who consume inadequate intakes of vitamin D, and when there is limited exposure to sunlight. Sometimes vitamin D deficient diets are associated with vegetarians and persons with milk allergy and lactose intolerance (Biser-Rohrbaugh and Hadley-Miller 2001). Patients who suffer from chronic intestinal malabsorption syndromes are more likely to develop vitamin D deficiency because the small intestine is unable to absorb this vitamin (Holick 2006) also parathyroid, liver and kidney disease can

negatively affect vitamin D status (Kalman 2006). In addition, several other factors can affect vitamin D status such as: obesity (Wortsman et al. 2000), chronic alcoholism, genetics, drugs, and gender differences (Collins and Norman 1991). The classical consequences of severe vitamin D deficiency are: rickets among infants, young children and adolescents (Chesney 2003), osteomalacia among adults (Eastwood 2003) and osteoporosis, which is present in older adults, postmenopausal women, persons who have difficulty walking or exercising and patients on long-term steroid therapy (LeBoff et al. 1999).

2.8.1 Hypovitaminosis D: A global issue

Vitamin D deficiency and insufficiency are globally still very common especially in risk groups such as young children, pregnant women, elderly and immigrants (Lips 2010). This nutritional deficiency has been recognized as a frequent problem in studies of different age groups in different countries. In addition, numerous data have shown marked seasonal differences as winter seasons may reduce the quality of sunlight of the appropriate wavelength for cutaneous synthesis of vitamin D. Table 2.2 shows the variation in serum 25(OH)D concentrations between different countries.

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Table 2.2 Prevalence of hypovitaminosis D among adults around the world

Reference	**Study population**	**Country & Latitude**	**Time**	**Age(y)** Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**Comments**
Gloth et al. (1995)	244 subjects	**USA**	-	≥65	30	-	Homebound elderly persons were likely to suffer from vitamin D deficiency
Chapuy et al. 1997	1569 subjects	**France** Twenty cities 43-51°N	Nov.- April	-	14% had ≤ 30 nmol/l	-	A high prevalence of vitamin D insufficiency in the general adult French population
Lamberg-Allardt et al. (2001)	126 males 202 females	**Finland** Helsinki-Vantaa & Turku-Loimaa area 60°N	Feb. -Mar. 1998	37.0±4.0 38.0±3.0	45.0±35.0 47.0±34.0	26.0±4.0 24.0±4.0	Vitamin D deficiency was common in the normal adult population in Finland
Rucker et al. (2002)	60 males 128 females	**Canada** Calgary	Winter Spring Summer Fall	63.8±11.9 64.3±12.7	57.3±21.3 62.9±28.8 71.6±23.6 52.9 ±17.2	26.7±3.9 27.3±6.5	A high prevalence of vitamin D insufficiency in study sample
Kudlacek et al. (2003)	400 males 648 females	**Austria**	Dec.-April	50.0±9·6 44·5±9·8	52.2±33.2	26·4±3·5 24·4±4·5	A high prevalence of hypovitaminosis D in study sample

Table 2.2 Prevalence of hypovitaminosis D among adults around the world

Reference	**Study population**	**Country & Latitude**	**Time**	**Age** (y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI (kg/m^2)** Mean ±SD	**Comments**
Oliveri et al. (2004)	113 males 226 females	**Argentina** seven cities 261-551°S	Aug.-Oct.	71.3±5.2	**North** 51.7±18.5 **Mid** 44.7±20.5 **South** 35.4±14.0	-	A high prevalence of hypovitaminosis D in the sample
Meddeb et al. (2005)	389 subjects	**Tunisia**	Jan.-Mar.	**Range** 20-60	47.6% had ≤37.5 nmol/l	-	The prevalence of hypovitaminosis D was increasing with age, and it was highly in females, multiparty, menopause and wearing the veil
Levis et al. (2005)	212 subjects	**USA** Miami	Mar. Sep.	54.6±13.1	**Males** 62.3±21.8 **Females** 56.0±20.5 **Males** 77.5 ± 27.5 **Females** 62.5 ±23.5	-	A high prevalence of hypovitaminosis D in the sample
Burgaz et al. (2007)	116 females	**Sweden** 60°N	Jan.- Mar.	**Range** 61-86	69.0±23.0	25.3±4.1	Dietary, supplementary intake of vitamin D, and taking a sun vacation were predictors for 25(OH)D during winter
Hintzpeter et al. (2007)	1763 males 2267females	**Germany**	Oct.1997 to Mar. 1999	**Range** 18–79	45.2 44.7	-	A high prevalence of vitamin D deficiency in the sample

Table 2.2 Prevalence of hypovitaminosis D among adults around the world

Reference	**Study population**	**Country & Latitude**	**Time**	**Age(y)** Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**Comments**
Binkley et al. (2007)	63 males 30 females	**USA** Honolulu Hawaii 21°N	Mar.	24.0±0.7	51% had <79 nmol/l	23.6±0.4	High amounts of sun exposure do not ensure what is currently accepted as vitamin D adequacy
Van der Mei et al. (2007)	1669 subjects	**Australia** Southeast-Queensland 27°S Geelong 38°S Tasmania 43°S	-	<60	67.0 75.5 51.1	-	Vitamin D insufficiency was common over a wide latitude range in Australia. Season appears to be more important than latitude
Orwoll et al. (2009)	1606 males	**USA**	Mar. 2000 Apr.2002	73.8±5.9	62.6±19.7	-	Vitamin D deficiency was common in older males and especially in obese, sedentary and living at higher latitudes in the USA
Garcia and Guisado (2011)	**Elite basketball players** 21 males	**Spain** Barcelona 41°N	April 2009 Mar. 2010	25.0±4.3	47.8±21.8	23.7±1.2	Professional basketball players were at higher risk of hypovitaminosis D after wintertime

Table 2.2 Prevalence of hypovitaminosis D among adults around the world

Reference	**Study population**	**Country & Latitude**	**Time**	**Age** (y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**Comments**
Horani et al. (2011)	151 subjects	**USA** California Orange County	-	-	19.2% had <74.9 nmol/l	-	A high prevalence of hypovitaminosis D in the sample
Nanri et al. (2011)	**Workplace A***	**Japan** Northeast Kyushu, 33.4-33.5°N	July	44.8±10.8	69.9±12.7	24.3±3.5	Vitamin D deficiency was common in Japanese workers during seasons with limited sunlight
	100 males			40.5±9.8	65.6±11.0	20.7±2.8	
	61 females						
	Workplace B[§]		Nov.	44.3±10.8	57.2±18.0	44.3±10.8	
	212 males			41.2±10.8	48.4±15.0	41.2±10.8	
	156 females						
Moy (2011)	158 male 222 female	**Malaysia** Kuala Lumpur	-	48.5±5.2	56.2±18.9 36.2±13.4	>80% were overweight or obese	Low 25(OH)D was associated with obesity, lifestyle and clothing style especially among females
Pablo et al. (2011)	40 males 42 females	**Argentina** Buenos Aires	Summer winter	-	**winter** 50% males 42.6% females had <49.9 nmol/l	-	Seasonal variation in 25(OH)D was observed with significantly higher concentrations in summer in both gender

***** Occupation (office work, %) 93.0 males, 55.7 females
[§] Occupation (office work, %) 87.3 males, 71.2 females

2.8.2 Hypovitaminosis D in the UK

Various studies have revealed that vitamin D deficiency is a serious problem for people living in the UK. This is because of several reasons including limited exposure to sunlight with the western lifestyle, as most work indoors during daylight hours and take little exercise outside combined with low dietary intake of vitamin D. Vitamin D fortification is required for few foods in the UK e.g. margarine, but milk is generally not fortified. Whereas in the United States and Canada fortified milk provides most of the vitamin D in diet (Calvo et al. 2005). Moreover, in the UK there is low skin synthesis of vitamin D in a cloudy climate (Engelsen et al. 2005) and during winter, since vitamin D cannot be synthesised between October and April in the UK, it is assumed that winter requirements are met from the store accumulated the previous summer (Macdonald et al. 2008).

The dietary habits of Somali people living in Liverpool were investigated with respect to food items containing vitamin D. The recruitment included 15 males and 45 females; with mean age 42 y (range 18–81y) and 10 children; 3 males and 7 females; with mean age 10 y (range 6–17y). The results indicated an infrequent intake of foods rich in vitamin D and this probably contributed to bone and muscle pain which was self-reported by 62.1% along with regular visit to the doctors for osteomalacia (confirmed by patient records), in 36% of the sample (Maxwell et al. 2006).

Hyppönen and Power (2007) determined the prevalence of hypovitaminosis D in the white British population. Serum 25(OH)D concentrations were measured in 7437 Caucasians who were born in England, Scotland, or Wales during the same week of March 1958. The results showed a high prevalence of hypovitaminosis D during winter and spring, when serum 25(OH)D

concentrations <25, <40, and <75 nmol/l were found in 15.5%, 46.6%, and 87.1% participants, respectively.

Roy and others (2007) found a high prevalence of low serum 25(OH)D concentrations among young UK South Asian (Pakistani origin) females living in Greater Manchester. The sample included 78 women; with mean age 29.2 y (range 18-36). The results showed that the majority of the subjects (94%) had mean 25(OH)D ≤37.4 nmol/l, and 6% had mean 25(OH)D ≤12.5 nmol/l.

In a study performed by Macdonald and others (2008) to investigate vitamin D status, 3113 Caucasian postmenopausal women (aged 54.8 y) were recruited. Serum 25(OH)D concentrations were 59.2 nmol/l and 49.2 nmol/l in autumn and spring, respectively.

Hirani and colleagues (2008) assessed serum 25(OH)D concentrations of 1160 subjects aged ≥65 y living in private households in England. The prevalence of vitamin D deficiency [25(OH)D ≤25 nmol/l] was 12% in males and 20% in females. Moreover, 57% of males and 62% of females were vitamin D insufficient, when a higher threshold of <50 nmol/l serum 25(OH)D concentrations was used to define vitamin D insufficiency.

Another study conducted by Hirani and others (2009), investigated the association of predictors of 25(OH)D in two UK surveys: the National Diet and Nutrition Survey (NDNS) and the Low Income Diet and Nutrition Survey (LIDNS). A valid serum 25(OH)D sample was obtained in 1297 and 792 participants from the NDNS and LIDNS, respectively. The researchers found that the NDNS participants who were not receiving benefits (n=1054) had a mean 25(OH)D of 50.1 nmol/l, which was higher than among NDNS participants receiving benefits (n=243) who had a mean serum 25(OH)D concentrations of

43.0 nmol/l and the LIDNS sample mean serum 25(OH)D concentrations were 46.5 nmol/l. Additionally, the season of taking blood, skin colour, dietary vitamin D and supplement intakes were significant predictors for all three samples.

A longitudinal study was conducted to compare the seasonal variation of serum 25(OH)D concentrations in postmenopausal women residing in Aberdeen (338 Caucasian females; mean age 61.7 y) and Surrey (138 Caucasian and 35 Asian; with mean age of 61.4 and 59.9 y, respectively). Mean serum 25(OH)D concentrations in summer/autumn were 53.3 nmol/l among Aberdeen Caucasian, 60.4 nmol/l among Surrey Caucasian and 25.8 nmol/l among Surrey Asia samples. In winter/spring the mean serum 25(OH)D concentrations were 40.4, 43.6 and 23.2 nmol/l, among Aberdeen Caucasian, Surrey Caucasian and Surrey Asia samples, respectively (Mavroeidi et al. 2010).

2.8.3 Hypovitaminosis D in Middle East regions

The Middle East is a region with abundant sunshine (12-42°N). However, numerous studies had demonstrated low serum 25(OH)D concentrations across all age groups (El-Hajj Fuleihan 2009), as a result of low vitamin D intake, and inadequate sunlight exposure due to conservative dress (Table 2.3).

Although the Arab Gulf countries have experienced a significant improvement in economic and health status, vitamin D deficiency is still prevalent among women and children, especially breastfed infants whose mothers have low vitamin D stores and who lack exposure to sunlight (Serenius et al. 1984, Molla et al. 2000, Dawodu et al. 2003, El-Desouki 2003).

Table 2.3 Prevalence of hypovitaminosis D among adults in the Middle East

Reference	**Study population**	**Country & Latitude**	**Time**	**Age**(y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**Comments**
Sedrani et al. (1983)	**Young** 26 males 33 females	**Saudi Arabia** Riyadh 24°N	Apr. May Jun.	**Range** 18-26	21±7.7 28.7±10	-	A high prevalence of low vitamin D status and it was due to avoid sunlight exposure (dress style), low dietary intake of vitamin D
	Elderly 13 males 11 females			62±13	9.0±3.2	-	
El-Sonbaty and Abdul Ghaffar (1996)	**Females** 50 veiled 22 non-veiled	**Kuwait** Kuwait city 21°N	-	**Range** 14-45	14.5±5 30.0±8.2	-	A high prevalence of vitamin D deficiency and osteomalacia among veiled Kuwaiti females
Fuleihan and Deeb (1999)	465 females	**Lebanon** Nabi-Shit 33.5°N	Aug.-Oct.	**Range** 15-60	28±35	**Range** 22.7-32.6	A prevalence of vitamin D deficiency was due to dress style and low dietary intake of foods fortified with vitamin D
Ghannam et al. (1999)	321 females	**Saudi Arabia** Riyadh	-	35.4±11.3	24.5±17.2	26.5±5.2	A high prevalence of vitamin D deficiency in healthy Saudi females

Table 2.3 Prevalence of hypovitaminosis D among adults in the Middle East

Reference	**Study population**	**Country & Latitude**	**Time**	**Age** (y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI (**kg/m^2) Mean ±SD	**Comments**
Gannagé-Yared et al. (2000)	99 males 217 females	**Lebanon** Beirut Bekaa valley	Jan. -Apr.	41.3±5.5 39.4±5.6	35.7±18.6 19.0±14.4	26.4±3.2 25.7±4.5	Low 25(OH)D in veiled females in rural. Rural males had the highest 25(OH)D despite their low vitamin D intake
Guzel et al. (2001)	**Females** 30 veiled 30 western style	**Turkey**	-	15.7 ± 6.13	82.6±39.9 134.5±68.1	-	Veiled females had low 25(OH)D
Mishal (2001)	22 males 124 females	**Jordan** Amman 31°N	Summer Winter	**Range** 18-45	**Summer:** **Males:** 43.8±5.2 **Females** **Western Style:** 36.7±6.1 **Head scarf:** 28.3±4.5 **Veil:** 24.3±5.8 **Winter** **Males:** 34.7±4.2 **Females** **Western Style:** 30.9±4.6 **Head scarf:** 24.4±3.9 **Veil:** 22.7±3.0	-	Dress styles covering the whole body, totally or nearly totally, had adverse effects on serum 25(OH)D concentrations

Table 2.3 Prevalence of hypovitaminosis D among adults in the Middle East

Reference	**Study Population**	**Country & Latitude**	**Time**	**Age** (y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**Comments**
Ghazi et al. (2004)	490 males 682 females	**Iran** Tehran	Summer Winter	**Range** 3-69	**Summer** Males 137.3±67.4 Females 72.4±72.4 **Winter** Males 59.9±44.9 Females 34.9±34.9	- -	Serum 25(OH)D for males during winter were lower than summer. In females there was no significant in the concentrations of the seasons and this can be attributed to patterns of the traditional clothing and lifestyle
Hashemipour et al. (2004)	715 males 495 females	**Iran** Tehran	-	**Range** 20-64	**Range** 12.5-150.0	**Range** 26.44-27.66	A high prevalence of vitamin D deficiency in Tehran. In young and middle aged females were significantly lower than the older group
Atli et al. (2005)	**Males:** 87 old age home 24 own home **Females:** 138 old age home 171 in own home	**Turkey** Ankara 40°N	-	 75.5±7.3 72.6±5.0 75.0±7.0 72.4±4.7	 93.8±72.1 157.7±107.3 61.9±74.1 103.0±97.3	 - -	Vitamin D deficiency was due to low exposure to sunlight and dress style

Table 2.3 Prevalence of hypovitaminosis D among adults in the Middle East

Reference	**Study population**	**Country & Latitude**	**Time**	**Age** (y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**Comments**
Arabi et al. (2006)	**Home-dwelling ambulatory** 157 males 286 females	**Lebanon** Beirut 33.5°N	-	74.1±5.08 73.4±5.2	28.2 24.0	27.2±3.9 30.5±6.5	A high prevalence of hypovitaminosis D in this study sample
Saadi et al. (2006)	**Females** 175 premenopausal 84 postmenopausal	**United Arab Emirates** Al Ain	-	37.5±9.5 58.3±8.9	24.3±10.4 27.3±11.2	29.2±6.3 30.9±5.4	Vitamin D deficiency was highly prevalent in study sample and appears largely attributable to insufficient sunlight exposure
Heshmat et al. (2008)	5232 subjects	**Iran** Tehran Tabriz Mashhad Shiraz Booshehr	-	**Range** 20-69	**43.5% Males** **37.1% Females had** 12.5< 25(OH)D ≤ 25	-	A high prevalence of vitamin D deficiency and it is more evident in Tehran, capital of Iran
Hosseinpanah et al. (2008)	**Free-living females** 245 postmenopausal	**Iran** Tehran	-	**Range** 40-80	73.0 ±62.3	-	3% of the sample had 25(OH)D <25 nmol/l and 37.6% had 25(OH)D between 25 and 50 nmol/l

Table 2.3 Prevalence of hypovitaminosis D among adults in the Middle East

Reference	**Study population**	**Country & Latitude**	**Time**	**Age** (y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**Comments**
Moradzadeh et al. (2008)	5329 subjects	**Iran**	-	**Range** 20-69	72.1% males 75.1% females were vitamin D deficient	-	A high prevalence of vitamin D deficiency in Iran
AlElq et al. (2009)	**Males** 100 age 25-35 100 age ≥50	**Saudi Arabia** Eastern Province	Feb.-May	**Range** 28.2±4.5 59.4±15.6	10% deficient 18% insufficient 12% deficient 25% insufficient	-	The prevalence of vitamin D deficiency among healthy Saudi males is between 28% to 37%
Niafar et al. (2009)	**Females** 300 postmenopausal	**Iran** Tabriz	Jan.-Apr.	63.41±4.64	**Range** 4-144	28.1±5.1	Low 25(OH)D among postmenopausal women in north-west Iran
Elsammak et al. (2010)	89 males 52 females	**Saudi Arabia** Eastern-region	Dec.-Mar.	30.0±8.5 31.0±7.2	25.4±11.7 24.7±11.2	27.1 26.0	A high prevalence of vitamin D deficiency among Saudi Arabs
Hamilton et al. (2010)	**Athletes** 93 male	**Qatar** Doha	-	-	**91%** had 25(OH)D< 50	-	Vitamin D deficiency was very common among healthy Middle Eastern male athletes

Table 2.3 Prevalence of hypovitaminosis D among adults in the Middle East

Reference	**Study population**	**Country & Latitude**	**Time**	**Age** (y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**Comments**
Hekimsoy et al.(2010)	119 males 272 females	**Turkey** Manisa 38.36ºN rural, urban, semi urban	End of winter	45.1±17.5 45.1±17.2	49.9± 38.7 38.0±28.8	-	Vitamin D deficiency was likely due to traditional dress in rural areas allowing less sunlight exposure to the skin
Mahdy et al.(2010)	**Health care professionals** 340 healthy volunteers	**Qatar** Doha	Jan. 2007 Jan. 2008	-	**Overall** 29.2 **Males** 34.1 **Females** 25.7	-	A high prevalence of vitamin D deficiency among health care professionals in Qatar
Al Anouti et al.(2011)	**University students** 70 males 208 females	**United Arab Emirates** Abu Dhabi	Oct. 2009 Apr. 2010	21.0 ± 4.6 20.8 ± 4.0	27.3 ± 15.7 24.2 ± 14.9	23.7 ± 4.2 23.2 ± 5.0	Vitamin D deficiency is a major public health burden among young Emirati adults, because of sun deprivation in a sun-blessed country.
Elsammak et al.(2011)	87 males 52 females	**Saudi Arabia** Dammam	Dec. 2008 Mar. 2009	30.0±8.5 31.0±7.2	25.2±11.5 24.7±11.2	27.2±2.3 26.2±2.4	A high prevalence of a vitamin D deficiency in this sample despite > 65% of subjects having adequate exposure to sunlight and > 90% reporting adequate intake of dairy products

Table 2.3 Prevalence of hypovitaminosis D among adults in the Middle East

Reference	**Study population**	**Country & Latitude**	**Time**	**Age** (y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**Comments**
Kaykhaei et al.(2011)	993 subjects	**Iran** Zahedan	-	-	85.2% deficient 9.5% insufficient	-	A high prevalence of a vitamin D deficiency in the population of Zahedan, a sunny area in southeast Iran
Mallah et al.(2011)	99 males	**Jordan** Amman	Nov.	29.0±9.7	44.5±10	25.5±3.5	Low 25(OH)D in females wearing head scarf or veil were high and (76%) of males and (90%) western style dressed females had 25(OH)D below the recommended
	201 females						
	13.3% western style			22.5±5.2	40.0±8.3	21.3±3.0	
	46.7% head scarf			33.9±14.9	31.3±6.3	24.9±4.5	
	7.3% veil			39.3±15.3	28.5±3.8	27.1±4.6	

2.8.4 Hypovitaminosis D among Middle Eastern immigrant groups

Hypovitaminosis D had been observed in numerous studies among Middle Eastern immigrant groups in several countries. The studies indicated that factors other than latitude influence serum 25(OH)D concentrations in this ethnic group including: season, limitations in sun exposure, low dietary and/or supplementary intake of vitamin D, body fatness and low physical activity levels particularly among females where cultural dress may contribute to lower sun exposure, in part, for lower serum 25(OH)D concentrations (Table 2.4).

Table 2.4 Prevalence of hypovitaminosis D among Middle Eastern immigrants in different countries

Reference	**Study population**	**Country**	**Time**	**Age** (y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**Comments**
Glerup et al. (2000)	**Females** 60 Arab veiled 9 Danish Muslim (non-veiled) 44 Danish control	**Denmark** Aarhus	-	 32.2±1.4 37.1±3.4 36.1±1.6	 7.1±1.1 17.5±2.3 47.1±4.6	 27.8±0.7 26.7±1.7 24.4±0.7	In veiled females, vitamin D deficiency was the result of limitation of sunlight exposure and a low dietary
Brock et al. (2004)	**Elderly** 34 Middle Eastern	**Australia** Sydney metropolitan area	-	20% were age >75 y	21.0±20.0	48% were BMI>35	Vitamin D deficiency associated with sun exposure, low dietary intake, reduce exercise and high % body fat
Holvik et al. (2004)	**Turkish** 87 males 101 females **Iranian** 108 males 91 females	**Norway** Oslo	Feb.-Nov. 2002	**25(OH)D <25** in: Males 39 y Females 37 y	 23.0% 45.5% 28.7% 45.1%	 - -	Vitamin D deficiency associated with season, dietary intake, BMI and education. A higher proportion of females than males had vitamin D deficiency

Table 2.4 Prevalence of hypovitaminosis D among Middle Eastern immigrants in different countries

Reference	**Study population**	**Country**	**Time**	**Age** (y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**Comments**
Erkal et al. (2006)	101 Germans **Turkish** 327 residents of Turkey 566 immigrants in Germany	**Germany**	Mar. 2000	**range** 16-69	68.4 40.6 38.1	-	Vitamin D deficiency was among Turkish nationals independent of weather they lived in Turkey or Germany, especially in veiled females
Hobbs et al. (2009)	**Arab-American Females** 22 non-veiled 45 veiled supplemented* 20 veiled non-supplemented	**USA** Dearborn, Michigan	Apr. 2007	 25-46.8 27-46 32.8-44.3	 14.4±33.8 10.0-28.8 5.0±17.0	-	Vitamin D deficiency was associated with dress style, vitamin D intake from fortified foods or supplements and education
Madar et al. (2009)	**Turkish** 25 mothers 25 infants	**Norway** Oslo and Drammen	Mar. 2004 to Feb. 2006	 26·8± 5·7 6·7±1.4 w	 26·1±14·1 37.0±38.3	27·2± 4·2	Maternal 25(H)D associated with vitamin D supplements and education
Pinelli et al. (2010)	39% males 542 Arab-American	**USA**	-	38±13	75% had 12.5 to <50 nmol/l 24% had 50 to <100 nmol/l	28.4±5.5	Vitamin D deficiency was associated with insulin resistant and glucose intolerance in males

* On the basis of vitamin D intake from supplemented food sources (milk or vitamin D-fortified orange juice) and vitamin D pills

2.9 Vitamin D Toxicity

Although excessive exposure to sunlight does not cause vitamin D poisoning, dietary supplements of high doses can be highly toxic (Gibney et al. 2003, Insel et al. 2006). Toxicity in normal adults requires intake of more than 1000 µg/d on a long-term basis (Vieth 2011). Over-consumption of oral sources of vitamin D in adults can lead to hypercalcalciuria and hypercalcaemia (Holick 1998). Initially this presents as increase urination and thirst, and if prolonged, the body deposits excess calcium in soft tissues, causing pain and organ damage. The other symptoms include severe depression, nausea, vomiting, poor appetite, constipation, weakness, weight loss, and increased risk of kidney stones (Chesney 1989, Holick 1998, Insel et al. 2006). Furthermore, Pentti and others (2004) demonstrated that too high of serum 25(OH)D concentrations might enhance prostate cancer development.

2.10 Conclusion

The present review recognises that vitamin D is a needed nutrient for efficient absorption of dietary calcium and is required to promote normal mineralization of bone, as it is an essential regulator of bone metabolism.

Vitamin D adequacy prevents rickets in infants, children and adolescents and osteomalacia and/or osteoporosis and increased fracture risk in adults. Additionally, it is thought to play a significant role to maintain optimal overall health by preventing a variety of diseases such as, cancer, diabetes, immune disorders and cardiovascular diseases. Obesity has been found to be linked with lower serum 25(OH)D concentrations most likely as a result of decreased bioavailability of vitamin D from different sources due to its deposition in body fat stores. Therefore, obese individuals need higher-than-normal dietary intakes, sun exposure or doses of vitamin supplements.

Vitamin D is naturally present in very few foods and in the UK only a few foods are fortified with vitamin D but it is available in dietary supplements. It is synthesised in the skin after casual exposure to solar ultraviolet B (UVB); radiation (290-315 nm). However, many factors affect UV radiation exposure and the synthesis including: season (time of the year), time of day, length of day, cloud cover, smog, skin melanin content, and sunscreen use.

Cutaneous vitamin D and that obtained from food and supplements are biologically inert, until it undergoes successive hydroxylations in the liver and kidney for activation.

It is well documented that low serum 25(OH)D concentrations have been observed in different countries. It is becoming a major worldwide health problem, especially in developing countries, more particularly, in the Middle East region. Furthermore, low serum 25(OH)D concentrations have been noted amongst Middle Eastern immigrant groups living in Europe; the most at risk appear to be women who cover skin and /or veiled. Common causes of vitamin D deficiency include a very low sunlight exposure, obesity, together with the infrequent consumption of foods containing vitamin D.

In summary, vitamin D is a fat-soluble vitamin and also a hormone best known for its role in maintaining blood levels of calcium. Inadequate vitamin D status has been associated with a wide range of diseases and conditions. Overweight and obesity appears to be associated with lower vitamin D status in several studies. Low vitamin D status based on low circulating concentrations of serum 25(OH)D, has been reported worldwide. However; there were limited studies that investigated risk factors that can affect vitamin D status within the human body in Middle Eastern people in the UK; yet there is reason to suspect a high rate of vitamin D deficiency. Given the importance of vitamin D for human health

and the lack of studies assessing the vitamin D status of this ethnic group in the UK the main aim of this research is to determine the prevalence of vitamin D deficiency and insufficiency in a group of Middle Eastern people living in the UK. Factors affecting vitamin D status such as dietary and/or supplementary vitamin D intake and lifestyle will be examined (See chapter 3). Moreover, energy and macronutrient intake according to weight status will be estimated and underreporting will be investigated (See chapter 4). Additionally, there are few studies that have examined seasonal variation in Middle Eastern group in the UK and changes of serum 25(OH)D concentrations after body fat loss. Therefore, the effect of seasonal variation over 3 months during a period of reduce daily sunlight (October-January) on 25(OH)D will be investigated and the relationship between serum 25(OH)D concentrations and body fatness of a study sample in the South West of the UK (Plymouth) will also be examined. Finally the effect of moderate loss of total body fat induced by diet and increased physical activity on serum 25(OH)D concentrations and insulin levels will be observed (See chapter 5).

CHAPTER 3: Vitamin D intake and other risk factors for vitamin D insufficiency in Middle Eastern people living in the UK: A comparison of cultural and ethnic groups

3.1 Introduction

3.1.1 Cultural Diversity: Food habits of the Middle East populations

The Middle East region is geographically defined as Bahrain, Egypt, Iraq, Jordan, Kuwait, Lebanon, Oman, Qatar, Saudi Arabia, Syria, United Arab Emirates, Yemen, Palestine, Iran, and Turkey (Omran and Roudi 1993). Like many other regions of the world, it is extremely diverse in its national, linguistic groups, and human or cultural geography. It is home to numerous ethnic groups, including: Arabs, Kurds, Persian, Turkoman, Assyrian, Berbers, Azeris, Nubian, Balouchi, Lurs, Circassians, Gilaki&Mazandarani, Armenians, Emiri, South Asian, Greeks, Afro-Arabs and European. And it is very varied when it comes to religions, including: Muslim, Christian, Jews, Copts, Zoroastrian, Bahai, Yazidi, Druze, Greeks, parsis, Hindus, Alawi, Maronites and Indigenous. Middle East region is varied in its physical geography, and climate (Kumaraswamy 2003). The cuisines of Middle Eastern countries are similar. Nevertheless, each culture group has clearly different eating habits, food favourites and food preparation method, because the diet of any culture is influenced by foods that are native to these countries. “Food habits are an aspect of culture in which personal, social and situational factors interplay” (Obeidat 2002).

The religious beliefs of Muslims forbid pork and alcohol intake, while Christian has no religious prohibition regarding those two items (Twaigery and Spillman 1989, Chaudry 1992). Lamb is the most widely eaten meat. Beef, poultry and fish are also common foods, and most dairy products are eaten in fermented forms such as yogurt and cheese, while milk is not widely consumed, unless in

desserts and puddings (Nolan 2007). Wheat, most commonly as bread and rice accompany each meal. Legumes, such as chickpeas, lentil, navy beans, black beans, fava beans and red beans are used in many dishes (Packard and McWilliams 1994, Mermelstein 1999, Almas et al. 2003). Vegetables and fruits are served often, usually raw or mixed in a salad. Additionally, they are used in many dishes (Nolan 2007).

Eating patterns in some Middle East countries have rapidly changed as a result of socio-cultural and economic factors such as religion, beliefs, food preferences, gender discrimination, education, women's employment, mass media, speedy urbanization and migration movement (Musaiger 1993). Within these changes, the traditional diet of Middle East populations, which was characterized by high-fibre content, and was low in fat, cholesterol and sodium, has changed to a "westernized" diet with high intake of energy-dense foods rich in fat, cholesterol, free sugars, sodium and low in dietary fibre. Increase smoking with sedentary life-style has led to obesity and diet-related chronic illness such as cardiovascular disease, hypertension, diabetes and cancer (Al-Isa 1995, Al-Nuaim et al. 1997, Ajlouni et al. 1998, Musaiger 2002, Galal 2003, Nasreddine et al. 2006, Esmaillzadeh and Azadbakht 2008, Madanat et al. 2008).

3.1.2 Food patterns and related diseases among Middle East immigrants

Many reports have been published concerning dietary intake and food patterns of immigrants from Middle East countries who have settled outside their region. Koçtürk (2004), indicated that a bicultural dietary pattern had emerged among immigrant Iranian women in Sweden and the major reasons for change were the price of foods, lack of home-country products, convenience and the preferences of children. An American study suggested that the eating and

smoking habits of Arab-American population in southeast Michigan may increase the risk for developing cardiovascular disease (Hatahet et al. 2002). On the other hand, Alevizos and others (2006) demonstrated that the traditional nutritional habits of immigrant Arabian pregnant women in Greece seem to provide all micronutrients in sufficient quantities.

3.1.3 Nutritional assessment (Food Frequency Questionnaire)

The Food Frequency Questionnaire (FFQ) is a dietary assessment method designed to collect long-term nutritional consumption, over months or years. FFQ is the most used technique in large epidemiologic studies of diet and health to investigate risk factors for nutrition-related diseases (Mann & Truswell, 2007; Insel et al. 2006).

Participants complete the FFQ themselves by reporting their usual frequency of consumption of each item from a list of foods for a specific period (Block 1982, Thompson and Byers 1994). Many Food Frequency tools incorporate portion size questions, or specify portion sizes as part of each question to assess relative or absolute nutrient intake (Block 1989, Thompson and Byers 1994). FFQ may either comprise, only a few food items or it may contain up to 200 items, the length of the list in part depends on the focus of interest (Gibney et al. 2002). FFQ has many advantages including inexpensive coding, can be self-administered and required a little time to complete (Block 1989, Thompson and Byers 1994). However, it depends on the participant's self-description of his or her diet. Furthermore, its ability to assess the nutrient intake of groups with a dietary pattern highly different from the food list is limited and its validity is highly dependent on the correct selection of portion size and nutrient content assumption for each food (Obeidat 2002).

3.2 Aim

To compare factors affecting vitamin D status and estimated dietary intake of vitamin D between different cultural and ethnic groups from the Middle East now living in the UK.

3.3 Objectives

The objectives of this chapter were to**:**

- Assess dietary intake of vitamin D, and detect any possible lower intakes in the sample.
- Explore the relationship between lifestyle and vitamin D intake.
- Establish the prevalence of overweight and obesity.

3.4 Ethical Approval

The human ethics committee of the Faculty of Science /University of Plymouth approved the study protocol. An information sheet and consent form was attached to the questionnaire to be signed by all participants.

3.5 Subjects and recruitment

Middle East (Iraqi, Iranian, Egyptian, Lebanese, Jordanian, Syrian, Palestinian, Yemeni, Saudi Arabian, Kuwaiti, and Turkish) volunteers were recruited by directly approaching them and by using networks within local community groups through the University of Plymouth, Plymouth City Council, Plymouth refugee's council, Iraqi Cultural Attaché-London and Middle Eastern shops. The participants were students, refugees and immigrants living in the United Kingdom for at least 1 month before the study. All were aged 18 years old and over. Information was obtained from the subjects who could not speak English via an interview with the help of a translator in the refugee's council. Stamped addressed envelopes were attached to the questionnaires that were posted to increase the response rate. So, the participant could easily and anonymously

return the questionnaire to the address at the School of Biomedical and Biological Sciences, UOP. The subject's participation in this project was entirely voluntary, they could withdraw at any time or stage from the study. Furthermore their names, contact details and their medical with nutritional information were kept confidential, only an alpha numerical code was written on the questionnaire. The code list was kept separate from the study notes and electronic data, so enhancing confidentiality. For the purposes of the study, participants have been referred to be that code in all of the calculations, measurements and reports.

Inclusion criteria of the participants were:

- Geographic area (participants living in the UK)
- Area of origin/previous residence (Middle East)
- Age (18 y and older)

3.5.1 Questionnaire Design

Food Frequency Questionnaires (FFQ) are designed and used as an instrument, to measure a participant's usual food intake, in large epidemiologic studies of diet and health. FFQ is used as a valid method to collect basic information about food intake in any survey all over the world such as: in the UK (self – completion questionnaire) was used in the low income diet and nutrition survey (LIDNS) in order to obtain nationally representative data on the dietary habits of a low income population (Food Standards Agency 2007). In the USA (Kant and Thompson 1997, Dixon et al. 2000, Heuberger et al. 2001, Bazzano et al. 2002, Shahar et al. 2003). in Canada (Bright See et al. 1994), New Zeland (Quigley and Watts 1997), Australia (Worsley et al. 2003), and also in Brazil (De Marins et al. 2001).

FFQ has been employed in many studies of food intake, and diet assessment (Rockett et al. 1995, Kabagambe et al. 2001, Kalantar-Zadeh et al. 2002) for

example; to evaluate vitamins intake in a selected group (El-Qudah et al. 2008), or to find relations between vitamin D deficiency and vitamin D intake (Maxwell et al. 2006), and a FFQ was used also to examine the association between vitamin D intake and risk of a specific disease (Pittas et al. 2006).

In our study a food and health questionnaire was designed with the objective of estimating vitamin D containing foods and other factors hypothesized to effect vitamin D status (Appendix 1).

A written consent form was attached with the questionnaire to be filled by all the participants (Appendix 2). A blank format was developed to collect contacts' information including: last and first name, postal address, phone number and the e-mail, as a willingness and eligibility to participate in the next stage of the study.

The questionnaire took approximately 20 minutes to complete and it was composed of three sections: Section 1 included information about self- reported chronic diseases which can lead to or as a result of vitamin D deficiency, history of vitamin D deficiency, medications use; reported family chronic diseases, cigarette smoking; amount of time spent outside for work or leisure; use of sunscreen or clothes for sun protection. Also in this section height and weight were requested to calculate Body Mass Index (BMI). Participants were asked to leave this part blank if unknown. The Body mass index (BMI) was determined where BMI = the weight in kilograms divided by the square of the height in meters.

Section 2 was about the dietary intake. This section had questions about consumption of vitamin D containing foods (see below) and alcohol (an alcohol unit chart was attached); and use of dietary supplements (including amounts

and frequency of vitamins and/or mineral intake). Seven categories ranged from “never” to “daily/more”.

Section 3 of the questionnaire was designed to collect demographic information, such as: level of education, nationality, religion and ethnicity.

Dietary intake

Dietary data was obtained from the FFQ part of the questionnaire. The FFQ was based in part on a previous FFQ that evaluated the consumption of vitamin D and calcium-rich foods in an adult Middle Eastern population (Gannagé-Yared et al. 2000). This contained dietary sources of vitamin D that related to the dietary habits of a Middle Eastern population. This FFQ needed to be modified since participants of our study may also be eating British foods. Therefore other vitamin D containing foods that are available and commonly eaten in the UK were added. Sources of these were identified from the UK national dietary survey as the main contributors of vitamin D in the UK, such as oily fish, margarine, fat spreads and some breakfast cereals, as these are fortified with vitamin D (NDNS 2003). The frequency of consumption and amount of foods that contain smaller amount of vitamin D were also requested (such as eggs and meat) because these foods may make a small contribution to vitamin D status if eaten in large quantities. For analysis purposes, all food frequencies were transformed into times per day. The daily frequencies were multiplied with standard portion sizes to calculate food intake in grams. Vitamin D estimations were carried out using standard food portion sizes (Crawley 2002). The individual food items were converted to an approximate average daily intake of vitamin D (µg) for each participant using The Composition of Foods (Food Standards Agency 2002).

The Reference Nutrient Intake (FAO and WHO 2002) for vitamin D is 5(µg/d). So considering this, individuals were categorised into three arbitrary groups based on their intake: high (>10 µg/d), medium (5-10 µg /d) and low (< 5 µg/d).

3.6 Statistical analysis

Data handling and analyses were performed using Excel (2007) and Minitab (version 15.0, Ltd, Coventry) programmes. Chi-square statistics were obtained to compare and to find associations among all the categorical variables. Results are given as numbers, percentages, with confidence interval, means and standard deviations for numerical variables.

The WHO-BMI standards (WHO 2009) were adopted to define obesity. The classification is as follows: underweight defined as a body mass index <18.5 kg/m^2; normal weight as BMI 18.5-24.9 kg/m^2; overweight 25-29.9 kg/m^2; and obese as BMI >30 kg/m^2.

The percentage of the sample in different weight categories were calculated out of the total sample and stratified by gender, age group, nationality, ethnic origin and family diseases. In addition, the significance of difference in the proportion of individuals classified as underweight, normal range, overweight and obese were studied in relation with other factors such as diet and occupation.

Statistical analysis of vitamin D intake included calculation of means, standard deviations and percentages. Comparison of mean gender differences in Vitamin D intake was reflected by Student's t-test. Estimated daily intake of vitamin D (µg) was presented as a frequency distribution and the values of low, intermediate and high were calculated lower and upper limits as cut-off according to (Al-Murrani et al. 2000). One way analysis of variance (ANOVA)

and the 95% least significant difference (95% LSD) post hoc test was used to indicate differences between groups.

Multiple regression analysis was used to examine the relationship between the independent variables, specifically: region of birth, ethnic origin and education on the dependent variable, vitamin D intake.

3.7 Results

More than 350 questionnaires were distributed. Only 246 questionnaires were collected, while 242 participants were included in the study, (69.1%), 167 males (age31.5± 8.7 y, range19-71y) and 75 females (age35.9 ± 11.0 y, range19-66 y). All of the 242 agreed to participate in the first stage of the study which is food and health questionnaire, and 152 agreed to participate until the end of the study. Collected questionnaires were disqualified if the age of the subject was under 18 years or if subjects did not originate from Middle East region; four questionnaires from Libya, Sudan, and Bangladesh were not included in statistical analysis, which was performed on the 242 subjects.

The sample size of 242 subjects was within similar studies in the literature, which have recruited even smaller sample sizes (see chapter 3).

3.7.1 Demographic characteristics of the subjects

The sample distribution by gender, age group, region of birth, ethnic origin, religion, educational level and occupation status are detailed in table 3.1.

The vast majority of the participants were living in Plymouth 88%, followed by London 12%.

There were more males than females in the sample: 69% and 31% respectively. About 44.2% and 39.6% of the subjects were young aged 30 to 39 and 19 to 29 respectively, 10.3% aged 40 to 49, 2.8% aged 50 to 59 and 2.8% were aged 60 and above.

Looking at the subject's region of birth, 65.2% was Iraq, 12.3% Iran, 8.2% Arab gulf countries (Saudi Arabia and Kuwait), 5.7% Levant, 3.7% Egypt, 2.8% classified as other and 1.6% was United Kingdom.

Among the subjects 46.2% were Arab, 40.4% Kurd, 5.7% Persian, 3.3% classified as other, and 2.0% Arab/Kurd and 2.0% were Turkoman.

There was quite large percentage 87.1% of Muslims among the sample compared with 5.3% did not admit their religion, so we classified them as other-unidentified, 4.5% Christian and 2.8% classified as other (Druze, Mandaean, Yezidi, and Muslim/Christian).

About a quarter of the sample 25.6% had a postgraduate qualification, 18.5% have bachelor level, and 16.9% classified as other, 14.4% have secondary education level, 13.2% have Diploma education certificate and 11.1% have primary education.

Occupation status was classified as unemployed, student and employed; the percentages of these three groups were 46.2%, 30.1% and 23.5%, respectively.

Table 3.1 Demographic characteristics of the sample

Variable	Males (n=167) n (%)	Females (n=75) n (%)	Total (n=242) n (%)
Age groups (y)			
19-29	74 (44.3)	22 (29.3)	96 (39.7)
30-39	68 (40.7)	39 (52.0)	107 (44.2)
40-49	18 (10.8)	7 (9.3)	25 (10.3)
50-59	4 (2.4)	3 (4.0)	7 (2.9)
≥60	3 (1.8)	4 (5.3)	7 (2.9)
Region of birth			
Iraq	102 (61.1)	56 (74.7)	158 (65.3)
Iran	25 (14.9)	5 (6.7)	30 (12.4)
Arab Gulf Countries*	16 (9.6)	4 (5.3)	20 (8.3)
The Levant **	9 (5.4)	5 (6.7)	14 (5.8)
Egypt	5 (3.0)	4 (5.3)	9 (3.7)
Other	7 (4.2)	-	7 (2.9)
United Kingdom	3 (1.8)	1 (1.3)	4 (1.6)
Ethnic origin			
Arab	65 (38.9)	47 (62.7)	112 (46.3)
Kurd	80 (47.9)	18 (24.0)	98 (40.5)
Persian	11 (6.6)	3 (4.0)	14 (5.7)
Other	6 (3.6)	2 (2.7)	8 (3.3)
Arab/Kurd	4 (2.4)	1 (1.3)	5 (2.1)
Turkoman	1 (0.6)	4 (5.3)	5 (2.1)
Religion			
Muslim	146 (87.4)	65 (86.7)	211 (87.2)
Other-Unidentified	10 (6.0)	3 (4.0)	13 (5.4)
Christian	8 (4.8)	3 (4.0)	11 (4.5)
Other ***	3 (1.8)	4 (5.3)	7 (2.9)
Educational qualifications			
Primary	20 (12.0)	7 (9.3)	27 (11.2)
Secondary	31 (18.6)	4 (5.3)	35 (14.5)
Diploma	25 (15.0)	7 (9.3)	32 (13.2)
Bachelor	25 (15.0)	20 (26.7)	45 (18.6)
Postgraduate	33 (19.8)	29 (38.7)	62 (25.6)
Other	33 (19.8)	8 (10.7)	41 (16.9)
Occupation			
Unemployed	82 (49.1)	30 (40.0)	112 (46.3)
Student	40 (24.0)	33 (44.0)	73 (30.2)
Employed	45 (26.9)	12 (16.0)	57 (23.6)

* Saudi Arabia and Kuwait
** Syria, Lebanon, Palestine, Jordan and Turkey
*** Druze, Mandaean, Yezidi, and Muslim/Christian
NB: Percentages are correct to one decimal place.

3.7.2 Health – Related indices

3.7.2.1 Gender

There was a significant difference between the genders for smoking and drinking alcohol. More males were smokers (*P*=0.0001), and more males drank alcohol (*P*=0.006). The variability is high as indicated by the 95% confidence interval (CI) in table 3.2. This study observed that self-reported disease occurrence was different between genders (*P*=0.001), with females reporting this more frequently (Table 3.2). Significant differences (*P*=0.004) were found between gender and sunlight avoidance, by using sunscreen (*P*=0.0001), and by clothing (*P*=0.001). Females were more likely to avoid sunlight (Table 3.2). No significant difference (*P*=0.443) in supplement intake between males and females was found.

Table 3.2 Health-related indices according to gender

Variable	Males (n=167) % (95% CI*)	Females(n=75) % (95% CI*)	Total % (n=242)	*P* value
Smoking	32.0 (24.9-39.0)	8.0 (4.1-18.5)	24.8	0.0001
Alcohol intake	26.3 (19.6-32.9)	10.7 (3.7-17.6)	21.5	0.006
Reported diseases**	12.6 (7.5-17.6)	30.7 (20.2-41.1)	18.2	0.001
Reported family diseases ***	16.8 (11.1-22.4)	34.7 (23.9-45.4)	22.3	0.002
Medications use	18.0 (12.1-23.8)	30.7 (20.2-41.1)	22.0	0.035
Sunlight avoidance	39.5 (32.0-46.9)	58.7 (47.5-69.8)	45.5	0.004
Sun protection /Sunscreen use	4.8 (1.5-8.0)	28.0 (17.8-38.1)	12.0	0.0001
Sun-protection /Clothing ****	62.3 (54.9-69.6)	84.0 (75.7-9.22)	69.0	0.001

* Confidence interval
******Liver disease, Kidney disease, Parathyroid disorder, Intestinal disorder, Milk allergy and lactose intolerance, Osteoporosis, Diabetes, Heart disease, Hypertension, Arthritis and Asthma
*** Arthritis, Cancer, Diabetes, Heart disease, Hypertension, Liver disease, Kidney disease, Hypercolessterolium and Multiple sclerosis
****Sunglass, Long sleeved shirt, Headscarf, Veil and Hat

3.7.2.2 Age group

The survey showed that the percentage of chronic diseases and medication use increased strongly (*P*=0.001) from 28.0% (95% confidence interval (CI) 10.3-45.6%) to 64.3% (95% (CI) 39.1-89.4%) and from 20.0% (95% (CI) 4.3-35.6%) to 85.7% (95% (CI) 67.3-104.0%), respectively, between age group 40-49 to 50 of age and above (Table 3.3).

The data stated differences (*P*=0.005) between age groups with sunlight avoidance. The age group of 30 to 39 reported high sunlight avoidance compared with other age groups. Among the entire age groups (Table 3.3) the highest percentage 58.9% (95% (CI) 49.5-68.2%) of sunlight avoidance was in age 30 to 39, while the lowest percentage of sunlight avoidance 20.0% (95% (CI) 4.3-35.6%) was in age group of 40-49.

Table 3.3 Reported diseases, medications use and sunlight avoidance among the sample according to age

Variable	19-29 (n= 96)	30-39 (n=107)	40-49 (n=25)	≥50 (n=14)	Total % (n=242)	*P* value
Reported diseases						
%	14.6	13.1	28.0	64.3	18.18	0.001
95% CI*	12.1-17.0	6.7-19.4	10.3-45.6	39.1-89.4		
Medications use						
%	17.7	17.8	20.0	85.7	21.9	0.001
95% CI*	10.0-25.3	10.5-25.0	4.3-35.6	67.3-104.0		
Sunlight avoidance						
%	37.5	58.9	20	42.9	45.5	0.005
95% CI*	27.8-47.1	49.5-68.2	4.3-35.6	16.9-68.8		

*Confidence interval

3.7.2.3 Region of birth

Table 3.4 reflects significant differences (*P*=0.005) in region of birth and drinking behaviour. The highest percentage 46.7% (95% (CI) 28.8-64.5%) of drinking alcohol was among the subjects who were born in Iran and the lowest percentage 5.0% (95% (CI) 0.0-14.5%) was for Arab Gulf countries.

Table 3.4 shows also significant differences (*P*=0.01) between region of birth and medications use. Highest percentage of medications use 26.7% (95% (CI) 10.8-42.5%) was among the subjects who were born in Iran, while the lowest was 10.0% (95% (CI) 0.0-28.6%) among the subjects who were born in Egypt.

Table 3.4 also represents significant differences (*P*=0.05) between region of birth and sunlight avoidance. The highest percentage of sunlight avoidance 53.2% (95% (CI) 45.4-60.9%) was among the subjects who were born in Iraq and lowest percentage 20.0% (95% (CI) 0.0-44.8%) was among the subjects who were born in other countries.

Table 3.4 Drinking behaviour, medications use and sunlight avoidance among the sample according to region of birth

Variable	Iraq (n=158)	Iran (n=30)	Arab Gulf (n=20)	The Levant (n=14)	Egypt (n=9)	Other (n=11)	Total % (n=242)	*P* value
Alcohol intake								
%	17.1	46.7	5.0	28.6	20.0	40.0	21.5	0.005
95% CI*	11.2-22.9	28.8-64.5	0.0-14.5	4.9-52.2	0.0-44.8	9.6-70.3		
Medications use								
%	23.4	26.7	15.0	14.3	10.0	20.0	21.9	0.01
95% CI*	16.7-30.0	10.8-42.5	6.0-30.6	0.0-32.6	0.0-28.6	0.0-44.8		
Sunlight avoidance								
%	53.2	30.0	35.0	28.6	40.0	20.0	54.5	0.05
95% CI*	45.4-60.9	13.5-46.4	14.0-55.9	4.9-52.2	9.6-70.3	0.0-44.8		

* Confidence interval

3.7.2.4 Ethnic origin

The results of the present questionnaire show that there was a significant difference (*P*=0.05) between ethnic groups in drinking habits. The highest percentage of alcohol drinking was 50% (95% (CI) 23.8-76.2%) in Persian group and the lowest percentage was 11.1% % (95% (CI) 0.0-25.6%) in group other (Table 3.5).

This study demonstrates difference (*P*=0.005) between ethnic groups in chronic diseases. The highest percentage of reported diseases was 33.3% (95% (CI) 11.5-55.0%) in a group classified as other, while the lowest percentage was 10.2% (95% (CI) 4.2-16.1%) in Kurd group.

Table 3.5 also indicates significant difference (*P*=0.001) between ethnic groups and reported family diseases. The highest percentage of reported family diseases 36.6% (95% (CI) 27.6-45.5 %) was in the Arab group and the lowest percentage was 5.1% (95% (CI) 0.74-9.4 %) in Kurd group.

Significant difference (*P*=0.005) was found between ethnic groups in medications use. The highest percentage of medications use was 42.9% (95% (CI) 16.9-68.8%) in Persian group and the lowest percentage was 11.2% (95% (CI) 4.9-17.4%) in Kurd group (Table 3.5).

No significant differences were found among ethnic origins in smoking habits, sunscreen use, and sunlight avoidance by clothing and supplement intake.

Table 3.5 Alcohol intake, reported diseases, reported family diseases and medications use among the sample according to ethnic origin

Variable	Arab (n=112)	Kurd (n=98)	Persian (n=14)	Other (n=18)	Total % (n=242)	*P* value
Alcohol intake						
%	17.0	24.5	50.0	11.1	21.5	0.05
95% CI*	10.0-23.9	15.9-33.0	23.8-76.2	0.0-25.6		
Reported diseases						
%	21.4	10.2	28.6	33.3	18.18	0.005
95% CI*	13.8-28.9	4.2-16.1	6.6-50.5	11.5-55.0		
Reported family diseases						
%	36.6	5.1	21.4	27.8	22.3	0.001
95% CI*	27.6-45.5	0.74-9.4	0.0-42.8	7.0-48.5		
Medications use						
%	28.6	11.2	42.9	22.2	21.9	0.005
95% CI*	20.2-36.9	4.9-17.4	16.9-68.8	2.9-41.4		

* Confidence interval

3.7.2.5 Religion group

A significant difference (*P*=0.001) was found between religion groups in drinking habits. It was the lowest among Muslim group 14.2% (95% (CI) 9.4-18.9%) and highest 84.6% (95% (CI) 64.9-104.2%) in the other- unidentified group (Table 3.6).

Table 3.6 also shows a difference (*P*=0.005) in sunscreen use among religion groups. The highest percentage of sunscreen use 38.5% (95% (CI) 12.0-64.9%) was in the other-unidentified group and the lowest percentage 9.5% (95% (CI) 7.4-11.5%) was among Muslim group.

No significant differences were found among religion groups and reported diseases, medications use, reported family diseases, smoking habits, sunlight avoidance by clothing and supplement intake.

Table 3.6 Drinking behaviour and sunscreen use among the sample according to religion

Variable	Muslim (n=211)	Other-unidentified (n=13)	Christian (n=11)	Other (n=7)	Total % (n=242)	*P* value
Alcohol intake						
%	14.2	84.6	54.5	71.4	21.5	0.001
95% CI*	9.4-18.9	64.9-104.2	25.0-83.9	37.8-104.9		
Sunscreen use						
%	9.5	38.5	18.18	28.6	12.0	0.005
95% CI*	7.4-11.5	12.0-64.9	0.0-41.0	0.0-62.1		

*Confidence interval

3.7.2.6 Occupation status

There was a difference (*P*=0.009) in alcohol intake according to occupation status. The highest percentage 35.1% (95% (CI) 22.6-47.5%) of the drinkers was among employed subjects, while lowest percentage 15.0% (95% (CI) 6.8-23.1%) was among student subjects (Table 3.7). A significant difference (*P*=0.034) was found in sunscreen use according to occupation. The highest percentage of using sunscreen 19.2% (95% (CI) 10.1-28.2%) was in student group and the lowest percentage 6.2% (95% (CI) 1.7-10.6%) was among unemployed group (Table 3.7). There were no significant differences among occupation status and other dependent variables.

Table 3.7 Drinking behaviour and sunscreen use among the sample according to occupation status

Variable	Unemployed (n=112)	Student (n=73)	Employed (n=57)	Total % (n=242)	*P* value
Alcohol intake					
%	18.8	15.0	35.1	21.5	0.009
95% CI*	11.56-26.0	6.8-23.1	22.6-47.5		
Sunscreen use					
%	6.2	19.2	14.0	12.0	0.034
95% CI*	1.7-10.6	10.1-28.2	4.9-23.0		

* Confidence interval

3.7.3 Weights, Heights and Body Mass Index (BMI)

Table 3.8 describes the sample distributed according to weight, height and BMI. Mean values for weight and height among the subjects were 76.6 kg and 174.0 cm and for males, 67.4 kg and 161.2 cm for females with mean BMI value for males and females of 25.3 and 26.0 kg/m^2, respectively.

However, the percentage of the obese males was 8.3%, while the percentage of the obese females was 21.1% (*P*=0.05). Furthermore, overall, 40.7% of the males were overweight compared to 22.7% of the females. Less than 5% of both sexes were found to be under weight. In general, the prevalence of the obesity among females 21.1% was higher than males 8.3% (Table 3.9)

Table 3.8 Reported anthropometric data

Variable*	**Males (n=145)**	**Females (n=66)**
Weight (kg)	76.6±11.2	67.4±15.7
Height (cm)	174.0±0.1	161.2±0.1
BMI kg/m^2	25.3±3.5	26.0±6.1

* Data are expressed as mean ±SD

Table 3.9 BMI distribution of the study sample according to gender

Variable*	**Males (n=145)**	**Females (n=66)**
< 18.5 (Underweight)	1.4	3.0
18.5-24.9 (Normal)	49.6	53.0
25-29.9 (Overweight)	40.7	22.7
≥ 30 (Obese)	8.3	21.1

*****BMI classification according to WHO criteria (2009)

The mean values of the body weight, heights and BMI in different age groups are represented in table 3.10. The mean body weight and BMI were increasing gradually then declining from the age of 50 y and over among males and from the age of 60 y among females, while the mean heights among males and females were the same. The highest mean of BMI in the males and females was in age group of 30 to 59 y. In addition, females in age 50-59 y were obese.

Table 3.10 Body weight, height and BMI of the sample according to age and gender

Age group	**BW** (kg)*	**Height** (cm)*	**BMI***
Males			
19-29	73.1±10.2	1.7 ±0.07	23.9±2.9
30-39	79.6±11.2	1.7 ±0.06	26.5±3.5
40-49	79.8±8.8	1.7 ±0.05	26.3±3.4
≥50	74.1±16.8	1.7 ±0.08	24.7±5.1
Females			
19-29	59.0±9.4	1.5 ±0.06	23.2±3.9
30-39	68.3±14.3	1.6 ±0.06	25.9±5.4
40-49	73.8±16.4	1.6 ±0.05	29.1± 7.4
≥50	76.8±25.9	1.5 ±0.05	30.5±9.9

*Data are expressed as mean ±SD

Significant differences were seen between BMI with age (*P*=0.005) and BMI with ethnic group (*P*=0.005). There were no significant differences between other variables and the trend towards overweight and obesity (Table 3.10).

Table 3.11 also shows that the prevalence of overweight and obesity in the sample was the highest in age group 30 to 39 y among different age groups, and in Arab origin among different ethnic groups. The results do not indicate significant differences between the BMI and other variables.

Table 3.11 Percentage distribution of the sample according to BMI classification based on age and ethnic origin (between the groups)

		BMI Classification (%)				
Variable	**n**	**Underweight**	**Normal**	**Overweight**	**Obese**	***P* value**
Age group						
19-29	78	0.9	25.1	9.5	1.4	0.005
30-39	96	-	19.4	18.5	7.6	
40-49	23	0.4	3.8	4.7	1.9	
≥50	14	0.5	2.3	2.3	1.4	
Total	211	1.8	50.6	35.0	12.3	
Ethnic origin						
Arab	96	0.9	20.8	16.6	7.1	0.005
Kurd	87	0.9	22.2	15.1	2.8	
Persian	11		4.3	0.9	-	
Other	17	0	3.3	2.4	2.4	
Total	211	1.8	50.6	35.0	12.3	

Figure 3.1 shows the BMI distribution of the sample by reported family diseases (*P*=0.001). The figure shows that 43% of the normal weight sample reported chronic family diseases. However, it is important to note that 2, 24, and 31% of the individuals on reported chronic family diseases sample were underweight, overweight and obese, respectively. No associations between BMI and other dependent variables were found.

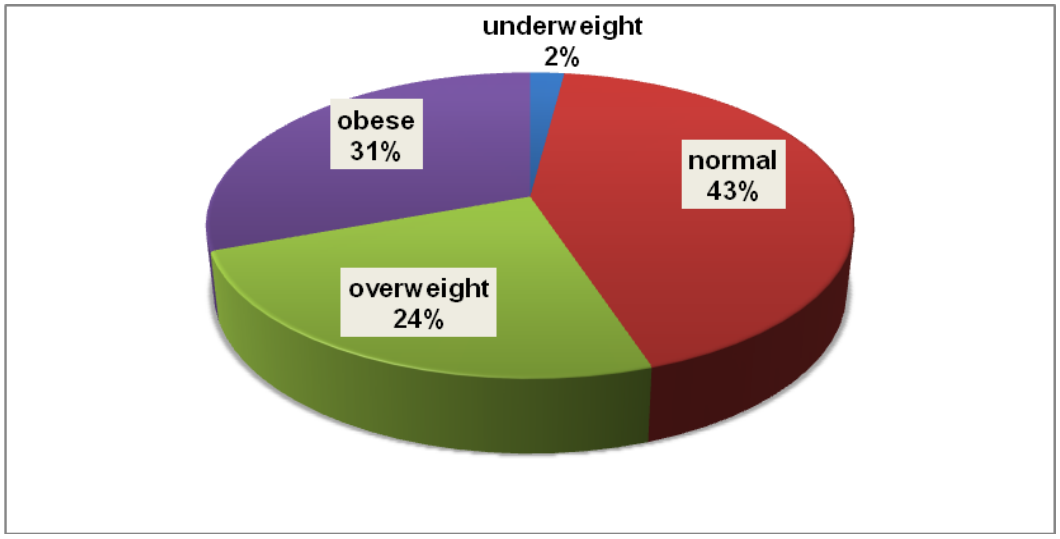

Figure 3.1 Percentage of BMI distribution of the sample who reported family diseases (n=49)

3.7.4 Vitamin D intake

Findings related to nutrient supplement intake among the sample; indicated that only 18.5% (95% (CI) 13.6-22.8%) of subjects were taking dietary supplements.

No significant differences were found between taking supplements and other variables. The frequency of intake of food groups which are rich in vitamin D among the total sample is presented in Figures 3.2 to 3.8.

3.7.4.1 Fish /Shellfish

Looking at the consumption of fish, it seems that between 90% and 62% of the sample reported no intake of mackerel, sardines, other, shrimp, salmon and tuna. Other reported seafood included mussels, carp, catfish, haddock, mullet, sea bass, tilapia and trout (Figure 3.2).

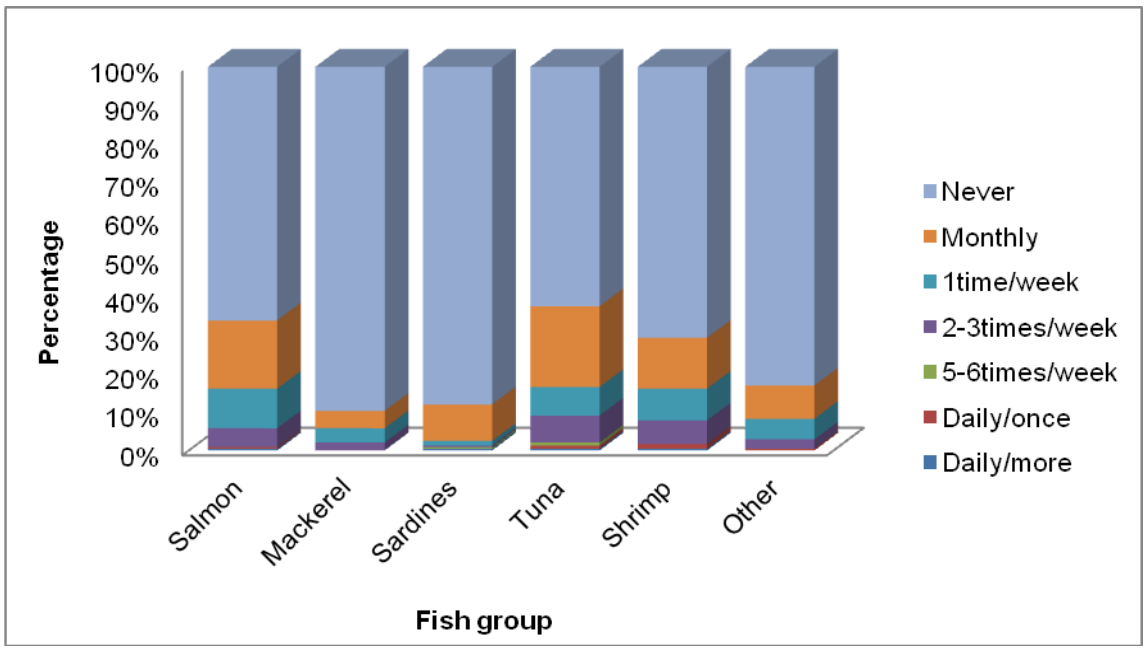

Figure 3.2 Frequency of consumption of fish group among total sample

3.7.4.2 Canned fish

Canned fish had low consumption rates, with 52%, 86% and 96% of the respondents reporting that they rarely or never eat tuna, sardines or other kind of canned fish, respectively (Figure 3.3).

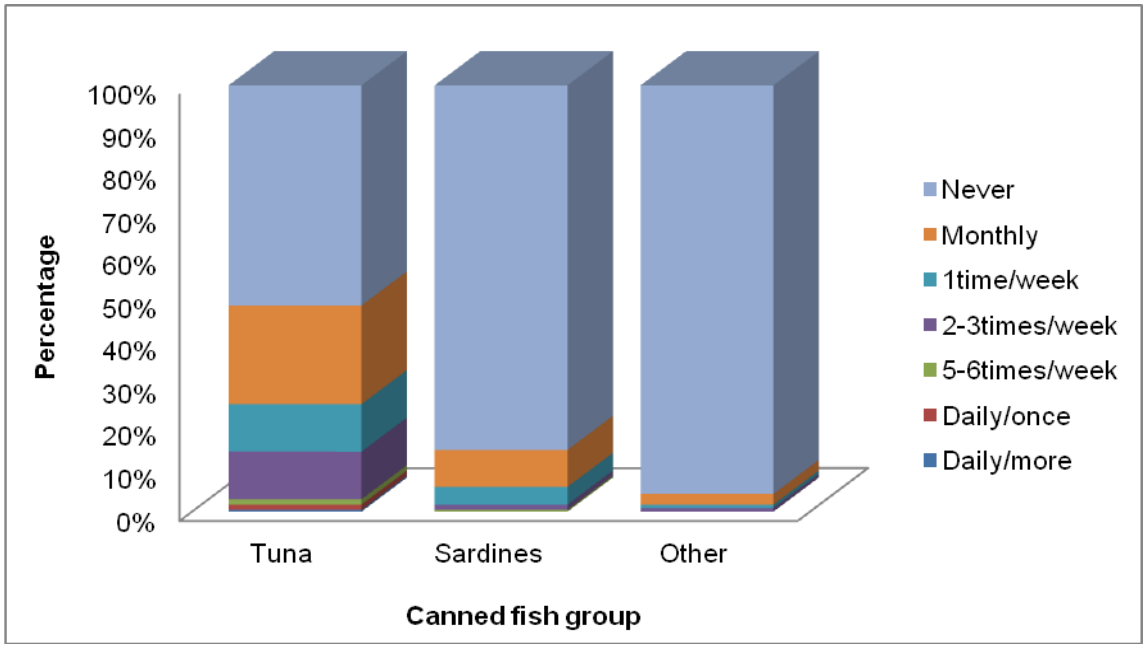

Figure 3.3 Frequency of consumption of canned fish among total sample

3.7.4.3 Meats group

For the meat group, 31% reported lamb consumption 2 to 3 times a week, followed by beef as 19% reported the same consumption frequency. However, 17% and 31% reported that they rarely or never had lamb meat, respectively. Whilst 44% reported that they did not eat burgers. Moreover, between (57-92%) reported that they never had goat meat, sausages, chicken, lamb, or beef liver. On the other hand, most rarely (95-99%) eat pork meat (Figure 3.4).

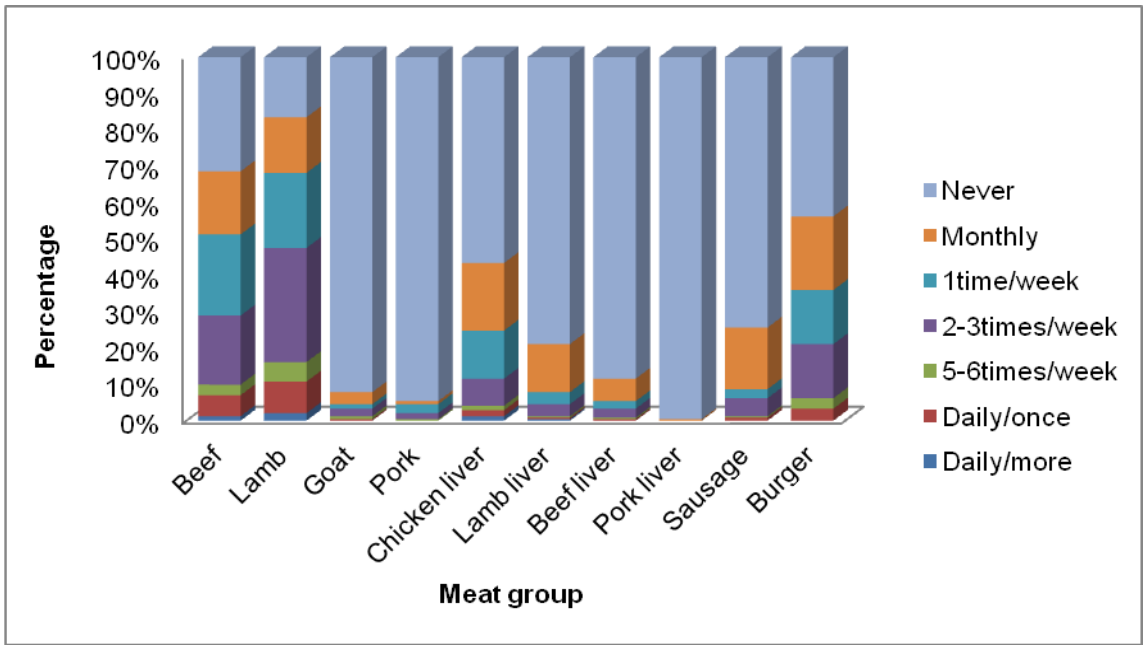

Figure 3.4 Frequency of consumption of meat group among total sample

3.7.4.4 Poultry

Chicken was consumed mainly 2 to 3 times a week as reported by under half of the subjects. On the other hand, turkey is not favoured, as 63% of the participants never eat it (Figure 3.5).

3.7.4.5 Eggs

Whole eggs are consumed mainly 2 to 3 times per week as reported by 29% of the sample compared with 3 % of yolks only and 2% of whites only, while 9% of

the subjects reported no intake of whole eggs compared with 91% of yolks only and 91% of whites only (Figure 3.6).

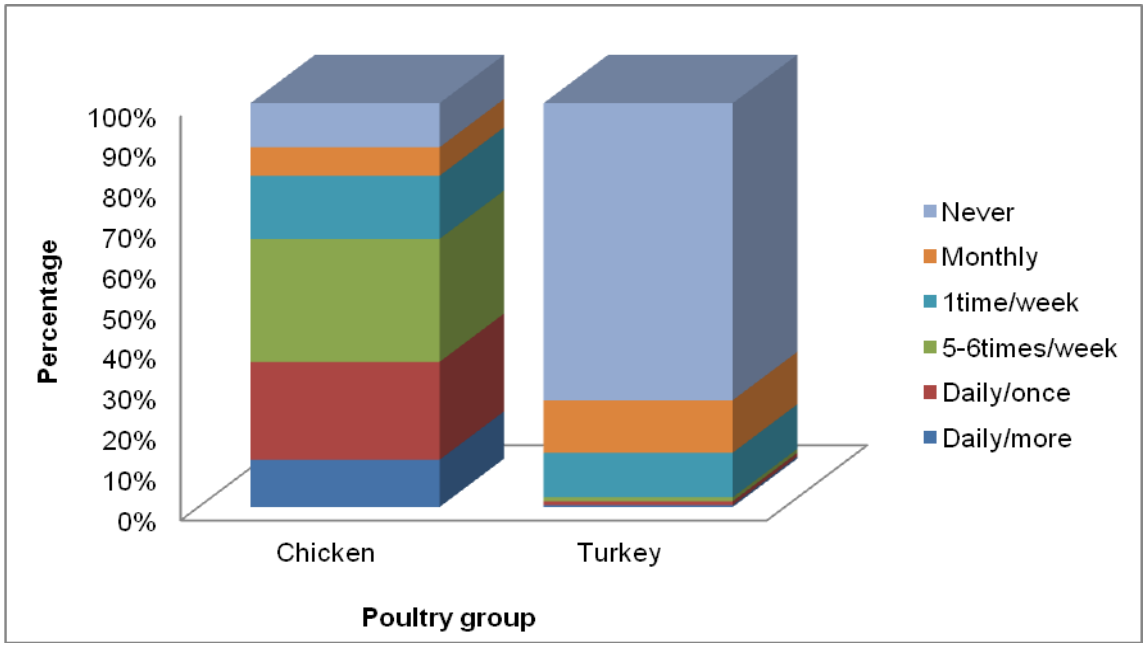

Figure 3.5 Frequency of consumption of poultry group among total sample

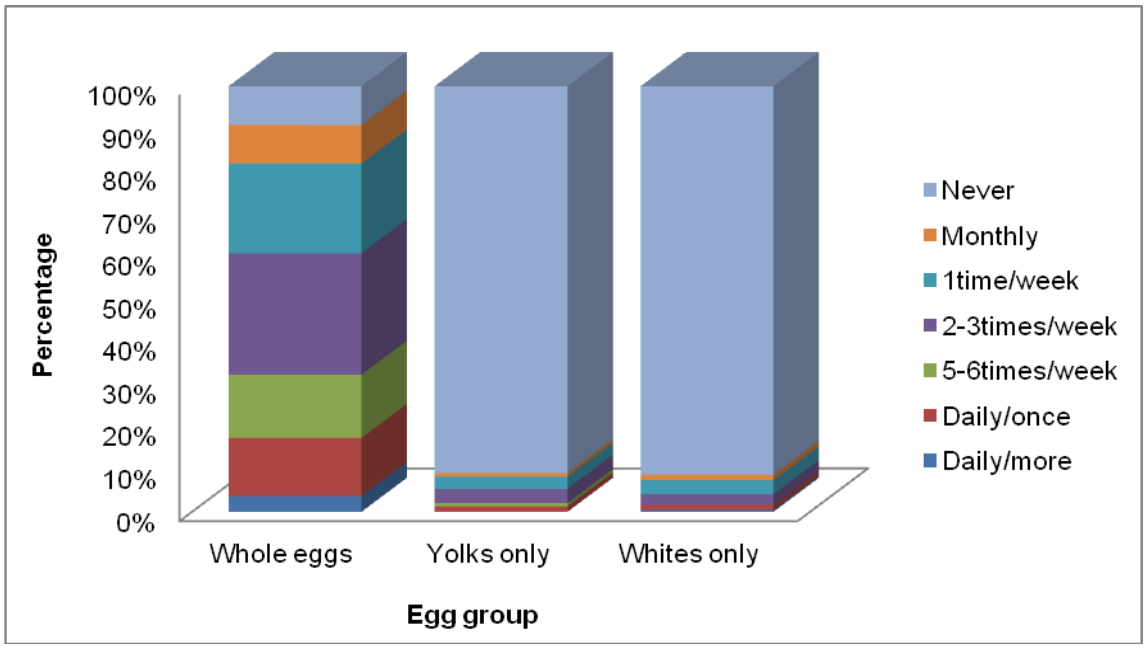

Figure 3.6 Frequency of consumption of egg group among total sample

3.7.4.6 Milk/ dairy products

Among the dairy products, cheese was the most popular being eaten 2 to 3 times a week by 37%, followed by yogurt 30%. Only 15% reported no intake for both two products. Additionally, 43% reported no intake of whole milk, 53% cream, 54% butter, 67% semi skim milk, 81% margarine and 83% skim milk (Figure 3.7).

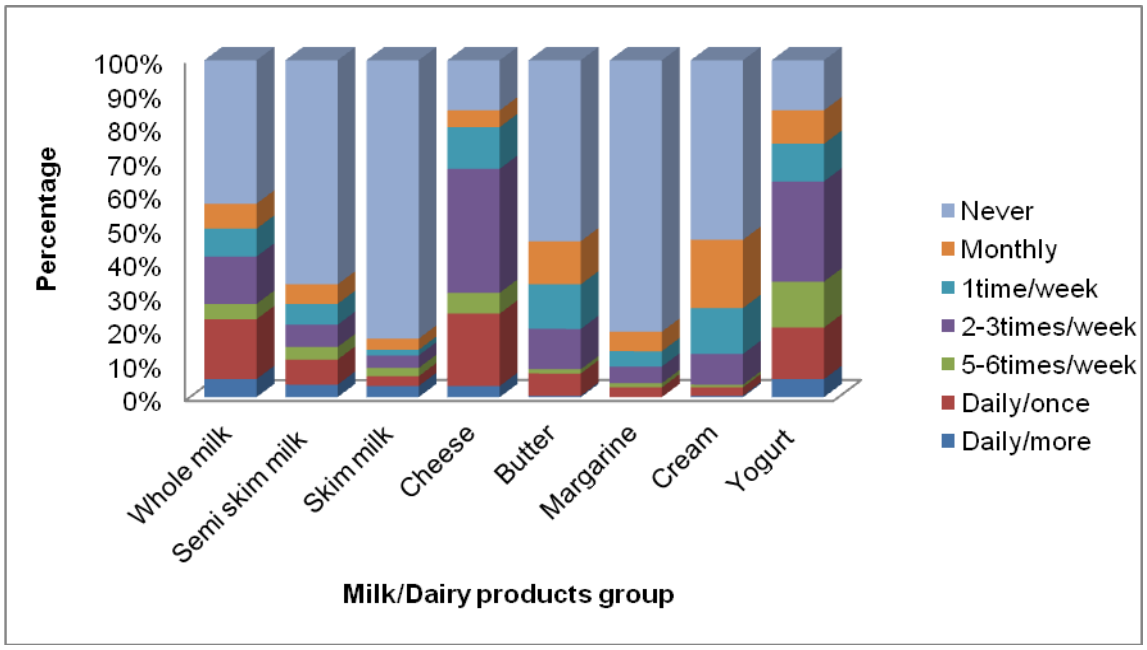

Figure 3.7 Frequency of consumption of milk/ dairy products group among total sample

3.7.4.7 Breakfast cereals

The consumption of breakfast cereals is relatively low (Figure 3.8), as 13% reported intake 2 to 3 times a week. Moreover 57% reported no intake.

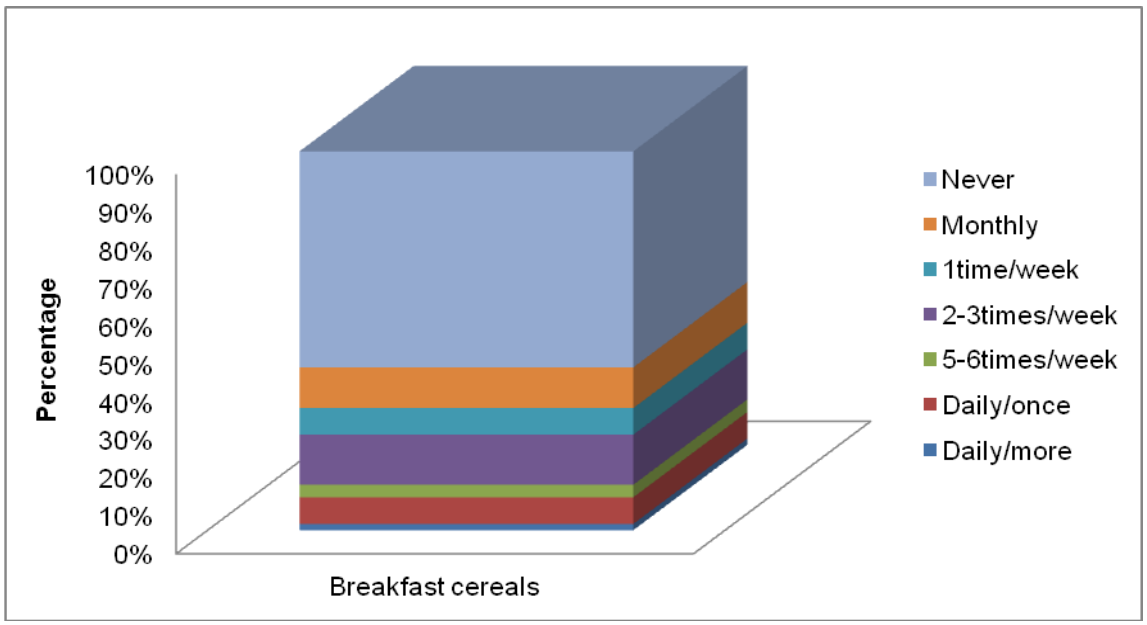

Figure 3.8 Frequency of consumption of breakfast cereals among total sample

3.7.5 Daily intake of vitamin D

In this study 85% of the sample had low intake of vitamin D which was less than 5 (µg/d), 12.6% had medium intake between 5-10 (µg/d) and 2.52% had a high intake which was more than 10 (µg/d) (Figure 3.9).

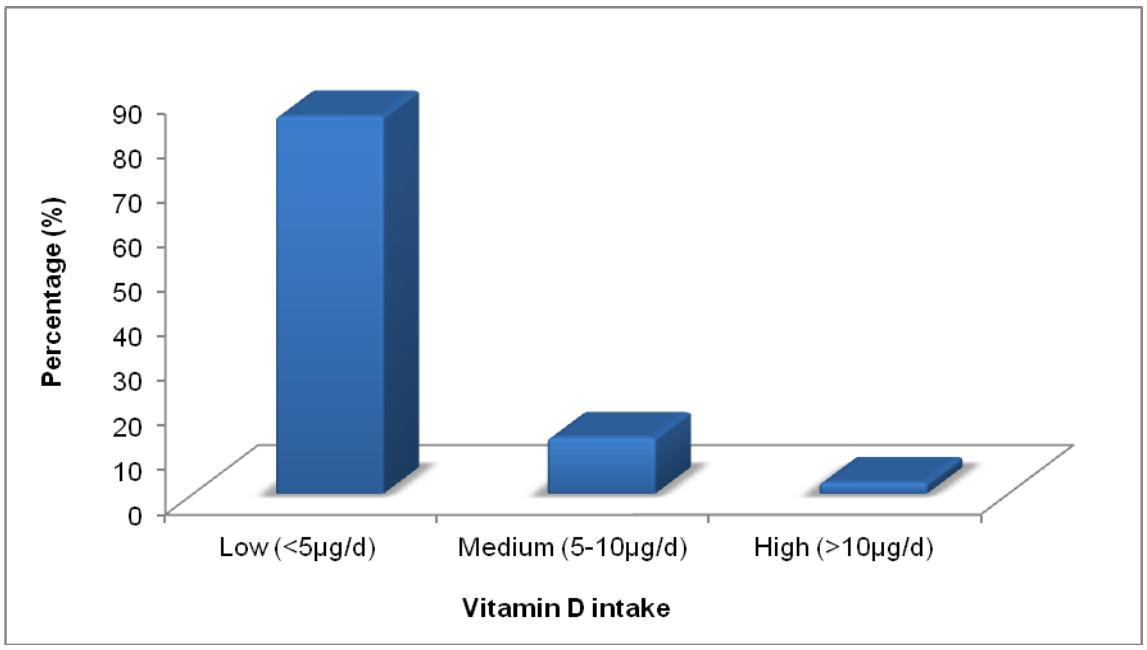

Figure 3.9 Percentage of vitamin D intake among total sample

The frequency distribution of mean daily intake of vitamin D was presented in figure 3.10. The 95% Confidence Interval (CI) and the lower and upper limits were calculated. It was found that 49.35% of the sample has a lower intake than 1.87 (µg/d), 6.06% consume higher than 7.62 (µg/d) and 44.56% in between. This distribution was constructed to further describe the estimated vitamin D intake for the sample.

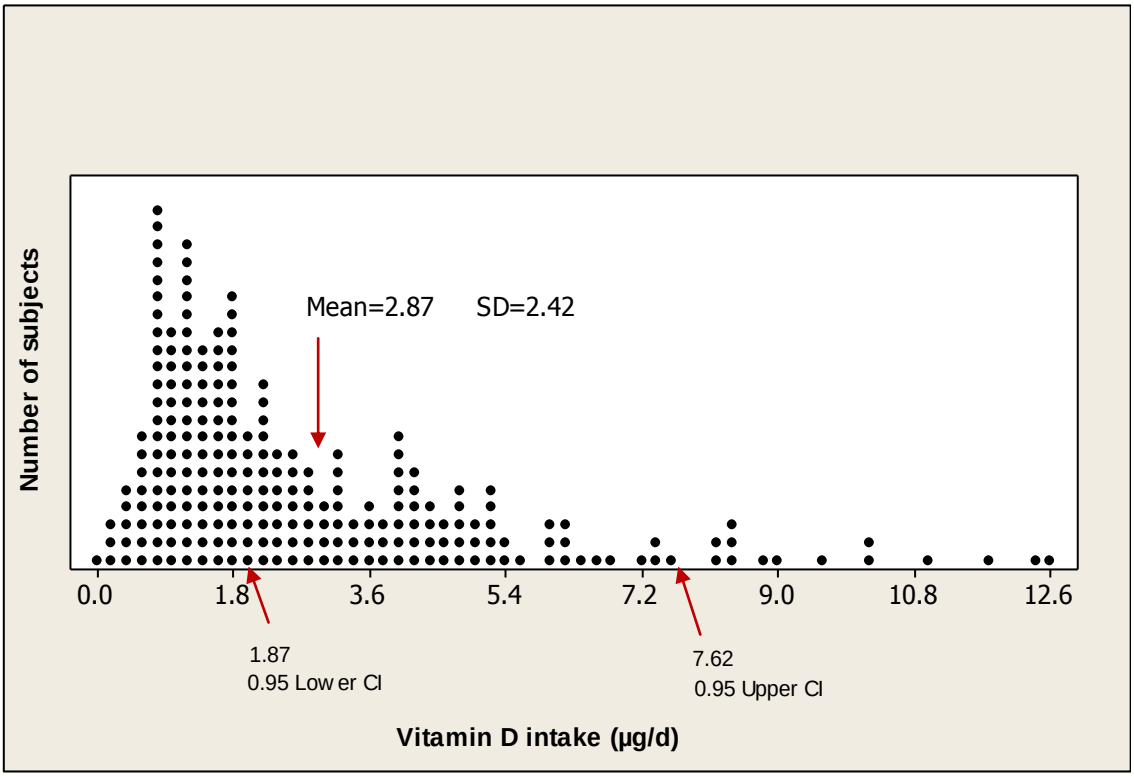

Figure 3.10 Distribution of estimated vitamin D µg/d, mean ±SD, upper and lower 95% confidence limits (CL)

The two sample t-test showed no significant differences of mean daily intake of vitamin D between males and females (Table 3.12).

Table 3.12 Estimated daily intakes for vitamin D according to gender

Gender	Vitamin D (µg/d) (mean ±SD)
Males	2.8±2.5
Females	3.0±2.2

T-test: no significant difference (*P* =0.621)

The one-way ANOVA showed no significant differences in mean daily intake of vitamin D (µg) according to the occupation status (Table 3.13).

Table 3.13 Estimated daily intakes for vitamin D according to occupation status

Occupation status	n	Vitamin D (µg/d) (mean ±SD)
Unemployed	112	2.7±2.2 [a]
Student	73	3.1±2.6 [a]
Employed	57	3.0±2.6 [a]

Post hoc test: LSD 5% =0.76, 1% = 1; no significant difference (*P*=0.411)

There were significant differences (*P*=0.027) of mean daily intake of vitamin D (µg/d) according to the participant's education levels. The highest mean (3.6 µg/d) was for bachelor level, while the lowest mean (1.8 µg/d) was for primary level (Table 3.14).

Table 3.14 Estimated daily intakes for vitamin D according to education level

Education level	n	Vitamin D (µg/d) (mean ±SD)
Primary	27	1.8±2.1 [a]
Secondary	35	2.1±2.0 [ac]
Diploma	32	3.0±2.0 [bc]
Bachelor	45	3.6±2.3 [b]
Postgraduate	62	3.2±2.9 [bc]
Other	41	3.0±2.7 [bc]

Post hoc test: LSD (Least significant difference) 5% =1.07, 1%= 1.41; means with the same letter indicate no significant difference. Any difference between two means carrying different letters is significant at 5% if it equals or exceeds 1.07 and at 1% if it equals or exceeds 1.41.

The one-way ANOVA reflected significant differences (*P*=0.05) in mean daily intake of vitamin D (µg) according to the age groups of the subjects. The highest mean (4.0 µg/d) was for age group of 40-49 y and the lowest mean (2.5 µg/d) was for age ≥50 y (Table 3.15).

Table 3.15 Estimated daily intakes for vitamin D according to age group

Age group	n	Vitamin D (µg/d) (mean ±SD)
19-29	96	2.8±2.7 [a]
30-39	107	2.7±2.1 [a]
40-49	25	4.0±3.0 [b]
≥50	14	2.5±1.8 [a]

Post hoc test: LSD 5% =0.94, 1%= 1.2; means with the same letter indicate no significant difference. Any difference between two means carrying different letters is significant at 5% if it equals or exceeds 0.94 and at 1% if it equals or exceeds 1.2.

Table 3.16 shows significant differences (*P*=0.003) between daily intake of vitamin D and the region of birth. The highest mean intake (4.3 µg/d) was among the participants who were born in region classified as the Levant and the lowest mean (2.4 µg/d) was among the subjects who were born in Iraq.

Table 3.16 Estimated daily intakes for vitamin D according to country of birth

Region of birth	n	Vitamin D (µg/d) (mean ±SD)
Iraq	158	2.5±2.1 [a]
Iran	30	3.3±2.8 [ab]
Arab Gulf	20	3.6±2.2 [b]
The Levant	14	4.3±2.9 [b]
Other	20	4.0±3.1 [b]

Post hoc test: LSD (Least Significant Difference) at 5% =1.14, at 1%=1.5; means with the same letter indicate no significant difference. Any difference between two means carrying different letters is significant at 5% if it equals or exceeds 1.14 and at 1% if it equals or exceeds 1.5.

The one-way ANOVA showed no significant differences (*P*=0.19) between mean daily intake of vitamin D and groups of sample's religion. However, Christian group seemed to have the highest mean daily intake of vitamin D (4.2 µg) but this was not statistically lower than other groups maybe due to the very small sample size of the Christian group. (Table 3.17).

Table 3.17 Estimated daily intakes for vitamin D according to religion

Religion group	n	Vitamin D (µg/d) (mean ±SD)
Muslim	211	2.8±2.4 [a]
Christian	11	4.2±2.8 [a]
Other	20	2.8±2.6 [a]

Post hoc test: LSD (Least Significant Difference):No significant difference.

Another significant difference found (*P*=0.001) was between mean daily intake of vitamin D and ethnic groups of the participants. The highest mean (4.2 µg/d) was for group other and the lowest mean (2.2 µg/d) was for Kurd group (Table 3.18).

Table 3.18 Estimated daily intakes for vitamin D according to ethnic origin

Ethnic origin	n	Vitamin D (µg/d) (mean ±SD)
Arab	112	3.2±2.4 [ac]
Kurd	98	2.2±2.2 [b]
Persian	14	3.2±2.3 [c]
Other	18	4.2±2.6 [a]

LSD 5%=0.93, 1% =1.2; means with different letters indicate a significance at 5%; means with the same letter indicate no significant difference. Any difference between two means carrying different letters is significant at 5% if it equals or exceeds 0.93 and at 1% if it equals or exceeds 1.2.

Table 3.19 presents significant differences (*P*=0.05) between mean daily intake of vitamin D and BMI values. The highest mean value of daily intake of vitamin D (3.6 µg) was for obese and the lowest mean (2.5 µg) was for underweight subjects.

Table 3.19 Estimated daily intakes for vitamin D according to Body Mass Index

BMI	**n**	**Vitamin D (µg/d) (mean ±SD)**
Underweight	4	2.5±1.3 [a]
Normal	107	2.8±2.5 [ac]
Overweight	74	2.6±2.4 [ab]
Obese	26	3.6±2.1 [bc]

LSD 5%=1.01, 1% =1.31; means with different letters indicate a significance at 5%. Means with the same letter indicate no significant difference. Any difference between two means carrying different letters is significant at 5% if it equals or exceeds 1.01 and at 1% if it equals or exceeds 1.31.

To examine the relationship between the independent variables, viz: region of birth, ethnic origin and education on the dependent variable, vitamin D intake, the regression analysis was used. The regression equation is: vitamin D intake =0.65+0.071 gender- 0.112 occupation status + 0.211 education level + 0.0158 age + 0.428 region of birth - 0.106 religion + 0.333 ethnic origin - 0.0089 BMI.

Looking at the regression equation; the regression values of education (+0.211), region of birth (+0.428) and ethnic origin (+0.333) were the only sizeable and positive effects that contributed to the overall significance. The inclusion of all of the above parameters in association with vitamin D in the regression analysis reflected a highly significant relation (*P*=0.004). This means that vitamin D is affected directly or indirectly by such parameters, namely: education level, region of birth and ethnicity.

3.8 Discussion

3.8.1 Vitamin D intake

The estimated mean vitamin D intake was 2.8 µg/d for males and 3.0ug/d for females which was not significantly different. A total of 85% of the sample had an intake of vitamin D which was less than 5 (µg/d). The results also showed that majority of subjects (81.4%) did not take vitamin supplements thus emphasising the importance of food sources of vitamin D for Middle Eastern people living in a country with no suitable UV radiation for subcutaneous synthesis during winter months.

In general, this study indicated that most of the participants do not favour fish; as between 62% and 90% of the participants reported no intake of fish/shellfish including oily fish. The subsequent chapters in this thesis have used repeat dietary recalls and a 3 day food diary to record food sources of vitamin D so it is useful to look at these results when interpreting the FFQ. These results are summarised in Chapter 6. The results of the subsequent studies also show a low intake of fish as out of 42 dietary assessments, fish (canned tuna) was only a source of vitamin D for one participant. A low consumption of fish was noted among adult urban population in Lebanon (Nasreddine et al. 2006). Moreover,Al-Khateeb and AL-Gelban (2008), were mentioned that fish is not served commonly among Saudi suburban community.

The results of the present questionnaire revealed that the intake of lamb meat was the higher compared with other kind of meat. This is corroborated by the subsequent studies in this thesis (Chapter 6) which showed that meat was the second main source of vitamin D for both the dietary recall method and food diary method, and lamb was the most important meat source followed by beef. These findings are in agreement with a report about food habits among Middle

Eastern emigrants in the United States, which reported that lamb is the most widely eaten meat (Nolan 2007). So although lamb meat is not particularly high in vitamin D compared to other foods, as it is eaten frequently it was the second most important source of vitamin D.

Regarding chicken intake, it was the most frequently type of meat among most of the subjects. Another study demonstrated similar result (Al-Khateeb and AL-Gelban 2008). However as chicken is lower in vitamin D (0.3 µg/100g compared to lamb 0.6 µg/100g) it was not such as important source to vitamin D intake in our other studies (Tables 6.1 and 6.2). Chicken was frequently eaten but lamb was a more important source of vitamin D for more people.

Results obtained from this study showed that whole eggs were widely consumed among the participants, while yolks or whites only were not consumed commonly. This was also seen in the subsequent nutritional analysis in this thesis (Tables 6.1, 6.2, 6.3) - eggs were commonly eaten and were the main source of vitamin D for over half of the nutritional assessments carried out.

This survey found that cheese and yogurt were the most common dairy products eaten. This result is in agreement with the report by Nolan (2007) who demonstrated that fermented forms of milk group such as yogurt and cheese are more common than other dairy products among Middle Eastern emigrants in the United States. In general this study indicated that the consumption of milk was low. In a study about food patterns of adults in the United Arab Emirates, the researchers reported that the consumption of milk was low (Musaiger and Abuirmeileh 1998). Another study of food consumption patterns in an adult urban population in Lebanon, Nasreddine and colleagues (2006) highlighted that the milk was the least consumed dairy products. In addition, milk

consumption was less than the daily requirements in a study to explore the wellness appraisal of Jordanian adolescents (Haddad et al. 2009). In the subsequent chapters in this thesis (summarised in Chapter 6) it was found that dairy foods (milk, yogurt, cheese and cream) did contribute to the total vitamin D intake and were the third most important source.

The present study indicates that around half of the participants do not consume breakfast cereals and consumption was found to be even less in chapter 6 where only 1 participant had a contribution to vitamin D intake from fortified breakfast cereals.

Therefore, considering the low consumption of vitamin D containing foods (such as: oily fish, fortified breakfast cereals and margarine) it was not surprising that approximately less than quarter of the sample had adequate vitamin D intake. From the results of this survey and the subsequent analyses in this thesis (Chapter 6) it can be seen that Middle Eastern immigrants to the UK are obtaining their vitamin D from eggs, meat (particularly lamb) and dairy foods. These foods do not have a high vitamin D content compared to fortified foods and oily fish, but do contribute as they are eaten frequently. The intake of eggs, lamb and dairy foods were not different according to region of birth, ethnicity and education level.

The later work (Chapter 6) confirmed that the most important dietary sources of vitamin D for Middle Eastern people in the UK had been included in the FFQ. In addition, the later work highlighted that some vitamin D rich foods widely available in the UK were included in the FFQ (mainly fortified foods and oily fish) but were not frequently consumed by this group.

A low intake of vitamin D will be expected to reflect on vitamin D presence in blood samples which will be taken in the next stage of the study.

The current study showed that there was no significant difference between the mean intake of vitamin D and the occupation status of the participants, while there were significant differences (*P*=0.027) between mean daily intake of vitamin D and the educational levels, the mean value of daily intake of vitamin D of diploma, Bachelor, postgraduate and other levels of education tend to be higher than primary and secondary levels.

In the present study, there were significant differences (*P*=0.05) between the mean daily intake of vitamin D and the age. Age group 40-49 y had the higher mean daily vitamin D intake than other age groups. In a survey carried out on behalf of the Food Standards Agency in the UK (2007), it was found that the mean daily intake of vitamin D among males was (3.3 µg), and among females was (2.5 µg), in age group 19- >65y, while in this study the mean daily intake of vitamin D among males was (2.8 µg), and among females was (3.0. µg) in age group 19->60 y.

The results of this study indicated that there were significant differences (*P*=0.003) between daily intake of vitamin D and region of birth. The subjects who were born in Iraq, Iran, and Arab Gulf countries had lower mean daily intake than the Levant and other countries. However, it should be noted that the Levant group only had 14 subjects and so this unlikely to be a representative sample of the region. Also the ‘other group’ is not a homogenous group and only had 20 individuals.

This study has shown that there were no significant differences (*P*=0.19) between mean daily intake of vitamin D and religion groups. Christian group

had the highest mean daily intake of vitamin D among the religion groups but again it should be noted that the Christian group was very small with only 11 participants and therefore is unlikely to be truly representative.

There were also significant differences (*P*=0.001) between mean daily intake of vitamin D and ethnic origin. The mean daily intake of vitamin D was the lowest in Kurd group and the highest in other ethnic group, while mean daily intake of vitamin D in Arab and Persian groups was in between. Here the sample contained large numbers of Arabs (n=112) and Kurds (n=98) which did have statistically different intakes (3.2 µg/d vs 2.2 µg/d respectively) but the Persian and other group were very small (n=14 and n=18 respectively) so were likely to be unrepresentative of the ethnic groups.

The results of this study found that there were significant differences (*P*=0.05) between mean daily intake of vitamin D and BMI values. Obese group had the highest mean daily intake of vitamin D among the BMI groups. This could be due to the fact that obese people eat more food and high-fat diets, which contain significant amount of vitamin D.

The regression coefficient (b) is positive and highly significant (P=0.004). The major contributor to the coefficient was the effect of the region of birth (b=0.428) followed by the ethnic origin (b=0.333) and the education (b=0.211). These variables were also significant in the univariate analysis. The overall picture indicates that there is a cause and effect relationship between those parameters and vitamin D intake.

3.8.2 The relationship between lifestyle and vitamin D intake

- **Smoking and drinking**

The prevalence of smoking and drinking was higher among males than females. A similar result has been reported by National Nutrition Survey in Bahrain (2002), Thom (2003) and Food standards Agency (2007) in the UK. This is likely to due to social and religious considerations, as it is less socially acceptable for women to smoke and drink, especially since 92.4% of our sample were Muslims. It has been found that smoking has a significant effect on vitamin D metabolism and smokers have significantly reduced levels of serum 25(OH)D concentrations (Brot et al. 1999). Moreover, alcohol consumption, especially if long term and heavy, might affect the metabolism of vitamin D; it has been observed that osteoporosis and osteomalacia are features of patients with chronic alcohol abuse particularly those with severe alcohol-induced liver disease (Lalor et al. 1986).

Another difference (P=0.005) was indicated between drinking and region of birth. Drinking behaviour was significantly the highest (46.7%) in those born in Iran and the lowest (5.0%) in those born in Arab Gulf Countries. Another study showed also significant differences between drinking behaviour as a social habits by region of birth (Balarajan and Yuen 1986).

The prevalence of drinking behaviour was the highest in Persians among other ethnic groups; numerous publications have stated that the likelihood of drinking has been associated with ethnicity and cultural diversity (Webb et al. 1996, Shatenstein and Ghadirian 1998, Denscombe and Drucquer 2000, Heim et al. 2004). It should be remembered that the number of Iranians and Persians was small in this study so we should treat these results cautiously.

Muslims are less likely than other religion groups to be drinkers; similar findings have been reported that the religion variables are important for drinking patterns (Raymond Cochrane 1990, Michalak et al. 2007).

In general, from the country of birth, ethnic and religion groups, this study indicated that drinking behaviour was high among Iranian subjects. Because Iran is a diverse country, it contains different ethnic groups, Persians form the majority of the population, and the main ethnic minority groups are the Azeris, Kurds, Arabs, Turkmen, Armenians and Assyrians. The religion held by the majority of the Iranian population is Islam. The remaining of Iranians are from 'other' religions – primarily Zoroastrian, Christian, Jewish, Bahai, Mandaean and Yezidi. The results revealed that there are differences (P=0.009) between drinking behaviour and occupation status. Employed subjects were more likely to be drinkers than underemployed or the students. (Montgomery et al. 1998); they also found that high alcohol consumption was significantly associated with recent unemployment.

- **Sunlight avoidance**

Females tend to avoid exposure to sunlight by using sunscreens or clothing more than males. This result is in conformity with the findings of Thieden et al. (2005), who indicated that females use sunscreens more than males. This might be due to the precaution taken by women to avoid sunburn and their concern about their skin and beauty more than men.

The subjects in age group of 30 to 39 were avoiding the exposure to sunlight more than other age groups. Moreover, no significant difference was found between the age and sunscreens use. Thieden and colleagues (2005) reported that sunscreen use is not correlated with age.

3.8.3 The prevalence of overweight and obesity

Overweight is of interest because it is associated with an increased health risks (Must et al. 1999, Kopelman 2000, National Task Force on the Prevention and Treatment of Obesity 2000) and it is associated with low serum concentrations of 25(OH)D (Wortsman et al. 2000). In this study, the obese group had a significantly higher vitamin D intake (3.6 µg/d) than the other BMI groups (2.5 - 2.8 µg/d) (*P*=0.05). This could be due to the fact that obese people have a higher total energy intake generally and more fatty food, which may contain vitamin D.

Results show high levels of overweight and obesity comparable to other populations. More males were overweight, while the prevalence of obesity was higher in females. Similar results were found in an adult in Jordan (El-Qudah 2008), in Lebanon (Sibai et al. 2003), In Bahrain (Bahrain MOH 2002), in Syria (Fouad et al. 2006), and in Palestine (Abdul-Rahim et al. 2003). Despite a higher intake of vitamin D in the obese group, excess body fat may be having a detrimental effect on vitamin D status by reducing its bioavailability.

The percentages of overweight including obese subjects in both sexes were the highest in the 30-39 y old age groups. While, Fouad et al. (2006) demonstrated that obesity increased with age being highest in age group of 46-65 among Syrian adults. Moreover, Food Standard Agency (2007) reported that the proportion of overweight and obese participants increased with increasing age. In this survey however there were small numbers of subjects in the older age categories and this might mean they were not truly representative of this age group. This survey discovered that the percentage of obesity was the highest in Arab group; this result is in conformity with the result of (Fouad et al. 2006).

3.9 Limitations

1- Recruitment of a compliant sample within the Middle East community in the UK was difficult. Recruitment was of a convenient sample and not a randomised selection. So the sample was limited to the groups available to the researcher which were postgraduate students and their families, those employed in institutions and companies known to employ Middle Eastern people and the refugee centre in Plymouth. This has limitations, for example: most of the females were postgraduate students or wives of postgraduate students and were highly educated. There were far fewer female refugees to interview who might have a lower education level and thus the sampling will have introduced some bias.

2- There was also likely to be selection bias based upon willingness to respond, some issues encountered were:

- Some people were extremely reluctant to participate in survey seeking personal information.
- Some people said they thought the questionnaire would take excessive time to fill it.
- A note from many subjects that they could not understand some of the items for example: sun protection factor; alcohol units, supplement; breakfast cereals, margarine, skim milk and yolk.

3- Some people could not read or speak English, so it was difficult for them to understand the context of the questionnaire; and for this reason some refused to sign the consent form, even one asked for a copy of the questionnaire for later reference.

4- The religious beliefs of Muslims forbid alcohol consumption. Therefore, it was embarrassing for some of them to declare it.

5- Many forms were not fully completed, so the subjects were contacted again by phone or meet them in their places of work or public places to follow up with interview and explain terms.

6- The FFQ was unvalidated - a suitable validated FFQ that included Middle Eastern and British foods for estimating vitamin D intake could not be found in the literature. So we used an unvalidated FFQ to base our FFQ on and then added British foods. The validity of the estimated vitamin D intake using this FFQ is considered in Chapter 6 when the results are compared to other methods of dietary assessment.

7- Also the food portion sizes were estimated from a food portion size guide for UK foods and consumers. So this might not be appropriate for Middle East foods. A food portion guide for Middle East foods was not available and so this limits the accuracy of the estimation of amounts of foods and therefore vitamin D eaten as Middle Eastern people might eat smaller of larger portions of these foods than in the typical British diet.

8- The demographic characteristics variables included small numbers and so must be interpreted with caution. Although, this is the first study to assess dietary vitamin D intake in Middle Eastern people living in the UK with a comparison between different demographic characteristics groups and therefore the detailed information is of particular interest. Every effort was made to convince as much as possible number of Middle East people living in Plymouth, Reading and London to fill in the Questionnaire. This study relied on accurate reporting and it is well known that height and weight and dietary intake are often misreported. However, a good response rate was achieved and the results show an overwhelming low intake of dietary intake of vitamin D plus the presence of other risk factors such as obesity and reduced sun exposure.

3.10 Conclusion

Risk factors that affect vitamin D status of Middle East people living in the United Kingdom aged ≥18 y were investigated. More than 85% of the sample was estimated to have an intake of vitamin D less than 5ug/d and less than quarter of the sample of participants took any vitamin supplements. Therefore the food sources of vitamin D will be of high importance to the vitamin D status of this group.

The participants in this study have low intake of fatty fish and other dietary sources of vitamin D such as margarine and other fortified foods. The main sources of vitamin D were from eggs, meat, particularly lamb, and dairy foods.

The present study found that there were no differences between mean daily intake of vitamin D and gender, between mean daily intake of vitamin D and occupation status and between religion group of the sample, whereas there were a significant differences between mean daily intake of vitamin D and other variables such as: education, age, region of birth, ethnicity and BMI. Moreover, vitamin D intake is affected directly or indirectly by education level, region of birth and ethnicity of the study sample.

A significant difference was found between gender with smoking habits, drinking behaviour, chronic diseases, reported family disease, medications use and sunlight avoidance. Males were more likely than females to smoke and drink. However, the percentages of chronic diseases, reported family diseases, medications use and sunlight avoidance among males were less; smoking habits and drinking behaviour among males could be attributed to different related chronic diseases and reported chronic diseases, reported family

diseases, medication use and sunlight avoidance among females increase the risk of vitamin D deficiency.

Another difference in the sample was recognized between age and chronic diseases, medications use and sunlight avoidance; the percentages of chronic diseases and medications use increased with the age, while sunlight avoidance was the highest in age groups 30-39 and ≥50 y.

Significant differences were discovered between region of birth and Drinking behaviour, medications use and sunlight avoidance; the highest percentages of drinkers were among subjects, who were born in Iran. Moreover, the highest percentages of using medications were among the participants who were born in Iran and Iraq, also the subjects who were born in Iraq were the highest percentage of avoiding sunlight exposure.

Ethnic origin had significant differences between drinking behaviour, reported chronic diseases, family diseases and medications use; Persian group tends to be more drinkers and use more medications among the different ethnic groups. This survey was found that percentages of chronic diseases and reported family diseases among the ethnic groups were the lowest in Kurd group and the highest in Arab group, respectively.

Religion had differences between alcohol intake and sunscreen use; Muslims tend to be fewer drinkers and they were the least people used sunscreen among the religion groups.

There were significant relationship between occupation status and alcohol intake and sunscreen use; employed subjects tended to drink alcohol more than

the unemployed and the students. However, students reported the highest use of sunscreen.

A significant difference was found between gender and BMI; males tended to be overweight and females tended to be obese. Highest percentages of overweight and obese among the sample were found within the age group of 30-39 year olds. In addition, over half of the participants who reported family diseases were overweight and obese. A significant difference was found between ethnic groups and BMI; highest percentages of overweight and obese were found in the Arab sub sample.

The results of this survey demonstrated that all Middle Eastern subjects may be at risk of vitamin D deficiency for many reasons including: their conservative dress limits their exposure to sun and they do not take significant amount of vitamin D from the diet or supplements. Biochemical assessment may be needed to confirm that there is low vitamin D status and based on that fortification can be considered, as a fortified food to boost the dangerously low serum levels of this vital substance.

CHAPTER 4: Dietary intake of vitamin D and calcium in Middle Eastern people living in the UK

4.1 Introduction

4.1.1 Measurement of food intake

The 24 hour dietary recall is a food record method based on amounts in detail of all the food and drink actually consumed by an individual during a period of time in the recent past (Lee and Nieman 2007).

It is an in-depth interview conducted by a trained dietary interviewer to obtain accurate quantification of amounts of foods about everything the subject had to eat and drink, from midnight to midnight of the previous day or over the 24-hour period, either backward or forward. It is quick and easy to administer (Eastwood 2003). The important factors in this method to obtain complete information are the use of open-ended questions, a non-judgemental manner/a neutral attitude /avoid asking questions in a manner that might influence the subject's responses, and the use of key questions and memory aids (Buzzard 1998).

In our study repeat 24 hour dietary recall has been chosen over other dietary assessment methods because it was shown by the Low Income Diet and Nutrition Survey in UK (Food Standards Agency 2007) to be more acceptable to low income groups and ethnic minorities than a food frequency questionnaire or food diary. There is less need to write English for people who have English as a second language and it is less burden for respondents than other methods.

4.1.2 Underreporting of habitual food intake

Lately, many publications have investigated the frequency of underestimation or measurement error in dietary assessment methods. Misreporting, particularly in the form of underreporting, is common but varies between research methodologies and sample populations (Heerstrass et al. 1998, Jonnalagadda

et al. 2000, Johansson et al. 2001, Ferrari et al. 2002, Kye 2004, Park et al. 2007). Under recording of energy intake has been a well-known problem in measuring food intake through self-reporting methodologies such as the 24-hour diet recall, especially among overweight people (Lichtman et al. 1992, Smith et al. 1994, Briefel et al. 1995, Heitmann and Lissner 1995b, Klesges et al. 1995, Ballard-Barbash et al. 1996, Fogelholm et al. 1996, Rothenbreg et al. 1997, Heerstrass et al. 1998) and women (Briefel et al. 1997, Mendez et al. 2004).

4.2 Aim

To obtain an estimate of energy and macronutrient intake of Middle Eastern people living in the UK

4.3 Objectives

The objectives of this chapter were to**:**

- Obtain a record of all food/drink participants consumed within proceeding 24 hours
- Estimate vitamin D and calcium intake
- Calculate BMI and relate to nutrient intake

4.4 Ethical Approval

The Human Ethics committee of the Faculty of Science /University of Plymouth approved the study protocol.

4.5 Subjects and recruitment

Participants of the study were recruited through the University of Plymouth and 28 subjects were included in this study. Out of a total of 14 females, 2 pregnant were excluded from the study. Participants were included from within the survey sample (See chapter 3). The research was carried out within the University of Plymouth campus (nutrition unit).

4.5.1 Anthropometric data

Height (cm) was recorded to the nearest 0.5 centimeter using a stadiometer (Model Seca 225, Hamburg, Germany) and weight (kg) was measured to nearest 0.1 kilogram using a balance scale (Model Seca, 880, Hamburg, Germany). Weight status was defined by calculating Body Mass Index (BMI) which is computed as body weight divided by height squared. The WHO-BMI standards (WHO 2009) were adopted to define obesity. The classification is as follows: underweight defined as a body mass index <18.5 kg/m^2; normal weight as BMI 18.5-24.9 kg/m^2; overweight 25-29.9 kg/m^2; and obese as BMI >30 kg/m^2.

4.5.2 Dietary recall

Dietary intake of subjects were obtained using the 24 hour recall technique on three random days (including one weekend day) within a 10 day period (Appendix 3). A written consent form was attached with the questionnaire to be filled by all the participants (Appendix 4).

Participants were given an explanation of the 24 hour recall: what it is, discuss purpose and what would happen with the information including their personal data.

The environment was made comfortable by keeping the desk neat, choosing a private and quiet location, away from distractions to help participants recall amounts/portions. It is easier for the participants to recall how much they ate when visual aids are used, and results will be more accurate. Therefore, the photograph method is recommended for the assessment of portion sizes (Lucas et al. 1995). Participants were asked to recall the food and drink consumed, food preparation methods, recipe ingredients, brand name of commercial

products, use of dietary supplements, how much was consumed, time it was consumed, how was it served, and specific information of the food (low fat, etc.).

Each participant kept the same alpha-numerical code that was given in the last study (Food and Health Questionnaire). This list was kept separate from the study notes and electronic data, so enhancing confidentiality. At the end of the study the list linking personal data to the study results will be destroyed.

To estimate portion size, both interviewer and each subject used household measures, which were converted to grams with the use of the photographic atlas of food portion Sizes (Nelson et al. 1997a) and food portion sizes : a user's guide to the photographic atlas (Nelson et al. 1997b). Energy and nutrient estimations were calculated using nutritional analysis programme (CompEat., 2003). For calculation purposes, Middle Eastern recipes were analysed to ingredient values when dealing with Middle Eastern dishes containing multiple ingredients. The CompEat programme does not contain traditional Middle Eastern dishes and therefore details of recipe ingredients were required from participants. Participants were asked to give verbally details of recipe ingredients. The proportion of each ingredient was estimated from the overall portion size eaten and this was entered manually.

4.5.3 Assessment of energy requirements

Basal Metabolic Rate (BMR) for each individual was calculated using Schofield (1985), prediction equations adopted by the FAO/WHO/UNU report (2004) based on age and gender (Table 4.1)

Table 4.1 Equations for estimated Basal Metabolic Rate (BMR)

Age (y)	Gender	BMR (kcal/d)	n	SE*
	Male	15.057 BW** + 692.2	2879	153
18-30	Female	14.818 BW + 486.6	829	119
	Male	11.472 BW + 873.1	646	167
30-60	Female	8.126 BW + 845.6	372	111
	Male	11.71 BW + 587.7	50	164
≥ 60	Female	9.082 BW + 658.5	38	108

*Standard error
**Body weight is expressed in kg

The ratios of the energy intake (EI) (kcal/d) /estimated Basal Metabolic Rate (BMR) kcal/d were calculated for each subject to evaluate energy underreporting. Subjects were classified as "energy under-reporters" when the ratio of reported energy intake to estimated Basal Metabolic Rate (EI/BMR) <1.14 according to the Goldberg *et al* (1991) cut-off limits. On the other hand, participants with EI/BMR >2.4 were classified as "energy over-reporters" based on the suggested range of the maximum energy for sustainable lifestyle which is between 2.0-2.4 (FAO/WHO/UNU 2004). Those with 1.14≤EI/ BMR≤2.4 were classified as "normal energy reporters".

4.5.4 Statistical analysis

Excel (2007) and Minitab (version 15.0, Ltd, Coventry) programmes were used for all the statistical calculations. Statistical methods included chi-square analysis for ascertaining differences between categorical variables; comparison of mean values ± standard deviation for gender and BMI differences in energy and nutrients intake was reflected by Student's T-test. Nutrient intake data were compared to UK Reference Nutrient Intakes (RNIs) (700 mg/d) and lower Reference Nutrient Intake (LRNI) (400 mg/d) for calcium (Department of Health 1991). Since the UK doesn't have RNIs for vitamin D (µg/d) in adults, FAO and

WHO (2002) recommendations were used: 5, 10 and 15 (µ/d) for age group of 19-50, 51-65 and >65, respectively.

4.6 Results

4.6.1 Age and Anthropometric data

Table 4.2 shows the anthropometric characteristics of the study sample. The mean age was 39.8±11.9 y (range 27-70 y) in males and 36.5±11.7 y (range 22-64 y) in females. A total of 58% of females reported sunlight avoidance by clothing. All of the subjects were from Iraq and the majority of them were students in the University of Plymouth and their families.

Table 4.2 Anthropometric data of the sample

Variable	Males (n=16)	Females (n=12)
Weight (kg)	83.3 ± 17.3	70.3 ± 10.3
Height (cm)	170.7± 7.9	158.4 ± 3.4
BMI	28.2±5.5	27.6±3.4

Data are expressed as mean ±SD

According to WHO-BMI classifications, both males and females lie in the overweight category. 16 (57.1%) and 6 (21.4%) subjects were overweight and obese, respectively. Figure 4.1 and 4.2 show the distribution BMI of males and females, respectively.

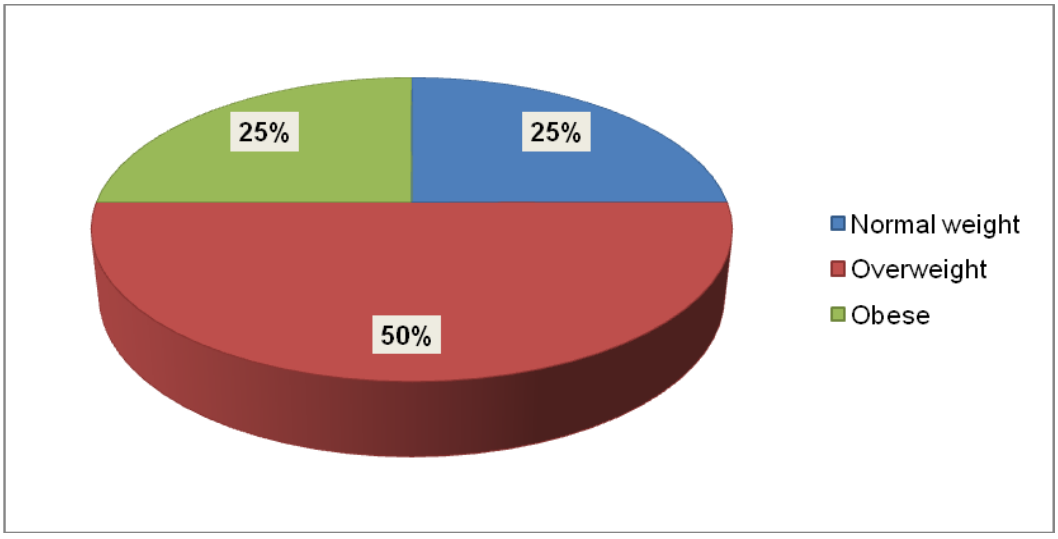

Figure 4.1 BMI of the Males

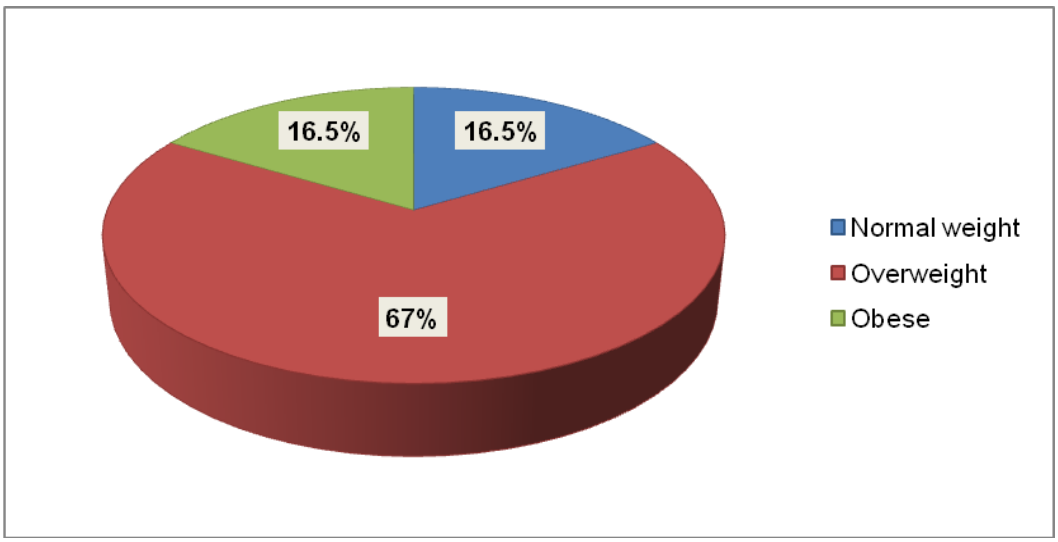

Figure 4.2 BMI of the Females

4.6.2 Energy and dietary intake

The two sample t-test showed no significant differences of mean daily intake of energy, protein, carbohydrates, fat and calcium between males and females. Whereas a significant difference (*P*=0.02) was found between males and females in dietary intake of vitamin D. The mean vitamin D intake recorded for males was (1.7±0.8 µg/d) and for females was (1.1±0.5 µg/d). However, inadequate intakes compared to the RNI were found for both calcium and vitamin D (Table 4.3). Although 53.6% of the subjects were below the Reference Nutrient Intake (RNI) for calcium, just 10.7% of the sample had intakes below the Lower Reference Nutrient Intake (LRNI) (the quantity sufficient for only 2.5% of the population). On the other hand, there is no LRNI for vitamin D; however, the entire sample had intake below the RNI (Table 4.4). The frequency of consumption of vitamin D rich food among the total sample is presented in table 4.5. Among all the food that contain significant amounts of vitamin D, eggs are the most commonly consumed followed by margarine, and supplement (containing vitamin D), whereas all the subjects in this study reported no intake for breakfast cereals and oily fish.

Table 4.3 Energy and nutrients intake of the sample compared to DRV* according to gender

	Males (n=16)		Females (n=12)		
Energy and Nutrients	**Mean ±SD**	**DRV**	**Mean ±SD**	**DRV**	***P* value**
Energy (Kcal/d)	1977± 575	2550	1586±556	1940	0.082
%Energy compared to recommendation for energy	77.5		81.7		
Protein (g/d)	78.2±24.1	55.5	66.8±20.2	45.0	0.186
% Energy as protein	15.6	15%	16.3	15%	
Carbohydrates (g/d)	255.9±70.5	-	203.8±71.6	-	0.068
% Energy as Carbohydrates	51.0	50%	49.8	50%	
Fat (g/d)	74.3±35.2	-	61.4±29.5	-	
% Energy as Fat	33.3	35%	33.7	35%	0.303
Calcium (mg/d)	672.0±231.0	700	696.6±242.0	700	0.788
Vitamin D (µg/d)	1.7±0.8	5**	1.1±0.5	5**	0.029

*Dietary Reference Values for Food, Energy and Nutrients for the UK (Department of Health 1991)
**RNIs (FAO and WHO 2002)

Table 4.4 Percentage below RNI /LRNI for calcium and vitamin D

Nutrients	% below RNI**	%LRNI*
Calcium	53.6	10.7
Vitamin D	All the sample	(no LRNI available)

* Reference Nutrient Intake (LRNI)
**The lower Reference Nutrient Intake (LRNI)

Table 4.5 Vitamin D rich food eaten

Vitamin D rich foods	Number of participants* consumed food	Total number of times in 3 days
Supplement (containing vitamin D)	1	1
Breakfast cereal (fortified types)	0	0
Margarine (fortified)	2	2
Oily fish	0	0
Eggs	28	41

* The number is out of 28 participants

4.6.3 Assessment of energy

From the study sample, 57.1% were under-reporters. In addition, a chi-square test was conducted to compare males and females for energy underreporting. The chi-square of freedom (x^2 1) showed no significant differences between males and females, although the rate of underreporting in males was 50%, while in females it was 66.6% (Table 4.6).

Table 4.6 Energy intake, expenditure and underreporting

	Males (n=16)	Females (n=12)
EI* (Kcal/d)	1977±575	1586±556
BMR** (Kcal/d)	1826±230	1414±109
EI/BMR ratio	1.1±0.36	1.1±0.36
Ratio of underreporting / Total (%)	50	66.6
Ratio of underreporting (overweight)/ Total (%)	58.3	80

Data are expressed as mean ±SD
* Energy intake
** Basal metabolic rate

For those who were overweight or obese, 68.1% of them were classified as under-reporters, while just 3.5% of normal weighted subjects were under-

reporters. Furthermore, the estimated ratios of underreporting were 58.3% and 80% among overweight males and females, respectively (Table 4.6). No significant differences for underreporting of energy were found between males and females by the level of BMI (Table 4.7). Furthermore no differences were found for mean daily intake of energy, protein, carbohydrates, fat, calcium and vitamin D between normal and overweight subjects (Table 4.7).

Table 4.7 Energy and nutrients intake of the sample according to Body Mass Index

Energy and Nutrients	**Normal weight (n=6)**	**Overweight (n=22)**	***P* value**
Energy (Kcal/d)	2038 ± 733.0	1747± 548.8	0.4
Protein (g/d)	71.3± 25.8	73.9± 22.5	0.83
Carbohydrates (g/d)	252.3 ± 91.2	228.4 ± 70.7	0.574
Fat (g/d)	80.3 ± 38.7	65.7± 31.5	0.428
Calcium (mg/d)	559.7± 139.7	716.0 ± 242.5	0.062
Vitamin D (µg/d)	1.3± 0.7	1.4± 0.8	0.776

Data are expressed as mean ±SD

The two sample t-test showed no significant differences (*P*=0.062) of mean daily intake of vitamin D between underreporters (1.2±0.6 µg/d) and acceptable reports (1.8±0.9 µg/d).

4.7 Discussion

4.7.1 Anthropometric data

This study found that more than half of the sample was overweight. A similar result was found in Middle Eastern subjects living in different cities in the UK in our previous study who reported their height and weight (Chapter 3, study 1).

4.7.2 Dietary intake

This study revealed a significant difference (*P*=0.02) between the mean daily intake of vitamin D and gender. Males had higher mean daily vitamin D intake than females. A similar finding has also been highlighted in a National Diet Nutrition Survey by the Food Standards Agency in the UK (2010). It was found that the mean daily intake of vitamin D among males was (3.1µg/d), and among females was (2.7 µg/d), in age group 19- 64 y while in this study the mean daily intake of vitamin D among males was (1.7µg/d), and among females was (1.1µg/d) in age group 19 to >60. This shows that the Middle Eastern group consumed lower amounts of vitamin D than the general UK population.

Overall, the mean daily intake of calcium and vitamin D in the sample were (682.5±231.6 mg) and (1.4±0.8 µg), respectively. The results of the dietary intake of calcium and vitamin D are similar to those of Gannagé-Yared and colleagues (2005) who used food frequency questionnaires to evaluate the consumption of vitamin D and calcium-rich foods in rural and urban Lebanese community centres. They indicated that mean daily calcium and vitamin D intake were respectively (683.8±281.2 mg) and (2.5±1.8 µg). Moreover, we demonstrated in chapter 3 that, Middle Eastern people living in the UK are thought to be specifically at risk of poor vitamin D status as a result of low consumption of vitamin D containing foods. The 24 hour dietary recall data also demonstrated low intakes of vitamin D fortified foods.

In this study more than half of the sample did not meet the RNI for calcium and none of the subjects met the RNI for vitamin D. Moreover, one tenth of the group did not achieve the lower Reference Nutrient Intake (LRNI) for calcium.

Mean daily intake of vitamin D was not significantly different between under-reporters and acceptable reporters. This is due to the fact that high fat snack foods are likely to be under-reported (Poppitt et al. 1998). Some of these foods (e.g. cakes, biscuits, pastries, contribute some vitamin D to the diet but they are not usually the main sources (see chapter 6).

4.7.3 Assessment of energy

The mean energy intake for the group was substantially lower than the UK estimated average requirement for energy intake which is likely to indicate underreporting.

The percentage of females underestimating energy intake was greater than males, although this was not statistically significant. This might be attributed to the small sample size and the high variability of the measurements. Similar findings have been also presented by other researchers (Johansson et al. 1998, Jonnalagadda et al. 2000, Johansson et al. 2001, Ferrari et al. 2002, Kye 2004, Yannakoulia et al. 2007). Women, compared to men, tend to be more preoccupied about weight, food and dieting; therefore, they are more prone to be embarrassed about their dietary intake and thus, more prone to be embarrassed about their dietary intake and thus, more prone to underreport (Macdiarmid et al. 1997). It seems this may also be true for Middle Eastern women.

In comparison to other groups where the 24 hour dietary recall has been used to estimate energy intake, the percentage of under-reporters in this study was

higher (50% males and 66.6 % females) than what was reported among a US population (18% males, and 28% females) (Briefel et al. 1997) and Swedish individuals (44% males and 47% females) (Johansson et al. 2001).

Our results indicate that, the percentage of under-reporters was higher in overweight and obese in both males and females compared to normal weight. These results seem to agree with the general assumption that overweight and obese individuals tend to underreport energy intake (Braam et al. 1998, Heerstrass et al. 1998, Johansson et al. 1998, Johansson et al. 2001, Ferrari et al. 2002, Park et al. 2007, Yannakoulia et al. 2007). Heitmann and Lissner (1995a) found that obese people tend to underreport fatty foods and foods rich in carbohydrates rather than underreport their total dietary intake. In addition, snack-type foods may be preferentially forgotten when obese people omit food items in dietary reporting.

So the observation that overweight and obese people under-report snack-type foods might explain why there was no significant difference in vitamin D intake between overweight / obese and normal weight in this study as snack type foods are less of an important contributor to vitamin D intake than eggs, meat and dairy foods (Chapter 6). However a different result was found in study 1 – where overweight and obese had a higher intake of vitamin D. So it could be that they are less likely to under-report with the FFQ, or it is due to the small sample size in study 2 and actually if there was a larger sample the difference could be significant. Or the FFQ is getting a longer term measure of vitamin D and we assigned portion sizes so these were not dependent on the individual. The recall was only over 3 days which recorded intakes for 3 days so provides only short term measures.

4.8 Limitations

1- Convenient sample of 30 participants was the maximum possible to allow reasonable data collection in time allowable.

2- The study sample was just one group (educated Iraqi subjects and their families) therefore not as representative of Middle Eastern people in the UK as our previous survey.

3- The estimation of dietary intake by using the 24 hour dietary recall, which contains considerable variations between one person to another, as not all of the sample were able to recall their food intake with enough accuracy.

4- Under recording of habitual food intake had effect on energy and micronutrients and in reality would be higher especially in the overweight group. Nevertheless, underestimation is more likely to affect certain meal types such as high fat snacks and thus is more likely to affect carbohydrate and fat estimations and less likely to have an effect on calcium and vitamin D results.

This is demonstrated by the comparison of vitamin D intake between under-reporters and adequate reporters – vitamin D intake was slightly less in the under-reporting group but it was not statistically significantly different to the adequate reporters.

4.9 Conclusion

In conclusion, underreporting of energy and nutrients intake is quite common among overweight and obese individuals. However, it does not mean that the subjects are dishonest about their food intake but may need more help in recording food intake accurately and a prospective method such as a food diary may be required. In the present study, all Middle Eastern subjects may be at risk of vitamin D deficiency due to limited intake of vitamin D from the diet or supplements, and overweight /obesity. Biochemical assessment is needed to confirm that there is low vitamin D status, and future work will look at the relationship between vitamin D status and body fat in this group.

CHAPTER 5: The relation between body fatness and vitamin D status of Middle Eastern people and Caucasians living in the South West of the UK

5.1 Introduction

5.1.1 Obesity and Health consequences

Obesity is a condition of abnormal or excessive fat accumulated in adipose tissue, to the extent that health may be impaired (WHO 2000). It is caused by imbalance between “energy in” (what is consumed through eating) and “energy expenditure” (what is used by the body) (Swanton 2008). Obesity has been identified as a risk factor for various diseases and illnesses including coronary heart disease, hypertension, stroke, type 2 diabetes, metabolic syndrome, osteoarthritis and cancer (Swanton 2008). Measures of obesity include: Body mass index (BMI), body fat, waist circumference and waist-hip ratio (WHO 2000).

5.1.2 Vitamin D and obesity

5.1.2.1 Vitamin D in fat tissue

Several studies have reported the relationship between vitamin D and body fat. The first evidence of this link was described by Lumb and colleagues (1971). They hypothesized that vitamin D, after absorption, is sequestered or stored in various tissues and then subsequently release slowly into the circulation and used biologically. In a subsequent study by the same team, (Mawer et al. 1972), they demonstrated that the highest concentration of parentally administered radiolabeled vitamin D, in 60 subjects, was seen in adipose tissue. Lawson and other researchers (1986) found a substantial amount of vitamin D in fat tissue samples taken from 15 British sudden death bodies.

Similar results have been found in rats (Rosenstreich et al. 1971, Brouwer et al. 1998). Such studies indicated that body fat compartments are the major storage site for vitamin D and a source available for conversion to other metabolites under fasting conditions.

Blum and others (2008b) reported that fat tissue and serum vitamin D concentrations were positively correlated, and that adipose tissue is a storage site for vitamin D.

5.1.2.2 Serum 25(OH)D concentrations and body mass index (BMI)

Body mass index provides a convenient way to estimate fatness. It is derived by dividing weight in kilograms by the square of height in meters (Grodner et al. 2012). BMI appears to be associated with lower serum 25(OH)D concentrations in several studies (Table 5.1).

5.1.2.3 Serum 25(OH)D concentrations and body fat

Low serum 25(OH)D concentrations have been found to be linked with body fat; Wortsman and colleagues (2000) demonstrated that obesity associated vitamin D deficiency is likely to be due to the decreased bioavailability of vitamin D from cutaneous and dietary sources as a result of its deposition in fat tissues. Lin and others (2011) found that vitamin D storage in adipose tissue was released during weight loss in 20 severely obese female patients who had Roux-en-y gastric bypass (RYGB). They concluded that serum 25(OH)D concentrations were increased over the period of rapid weight loss. Worldwide there have been several studies that have confirmed that body fat is associated with lower concentrations of serum 25(OH)D (Table 5.1).

5.1.2.4 Serum 25(OH)D concentrations D and waist circumference

Waist circumference is a measure of the accumulation of total body fat around the waist (NOO 2011). Higher levels of body fat around the waist is more dangerous than fat in the buttocks and thighs because it is associated adverse

changes in metabolism. Fat located in the abdominal area is called visceral fat and it related to health risk (Grodner et al. 2012). In several studies, central or abdominal adiposity may present a high risk to low serum 25(OH)D concentrations, as significant negative correlations were found between serum 25(OH)D concentrations and waist circumference (Table 5.1).

5.1.3 Vitamin D and insulin secretion

Recently, there has been increasing evidence from different studies, suggesting that vitamin D may play a potential role in modifying risk of diabetes.

Vitamin D has a role in pancreatic beta-cell function and this might be mediated by the binding of circulating $1,25(OH)_2D$ to the beta-cell vitamin D receptor (Pittas and Dawson-Hughes 2010). As an alternative, the function of vitamin D could be through activation of 25(OH)D by 1-alpha-hydroxylase, which is expressed in beta cells (Bland et al. 2004). The direct role for vitamin D to enhance insulin sensitivity might be via the stimulation of the expression of insulin receptors and/or by activating peroxisome proliferator activated receptor (PPAR-δ), a transcription factor that plays important roles in the regulation of fatty acids metabolism (Pittas et al. 2007). The indirect role for vitamin D to affect insulin secretion and sensitivity might be through regulating extracellular calcium concentration and fluidity across cell membranes in the beta cell and peripheral insulin-target tissues (Pittas and Dawson-Hughes 2010).

Several studies have investigated the link between vitamin D deficiency and insulin resistance and the important role of 25(OH)D to regulate type 1 and type 2 diabetes. A research study by Chiu and colleagues (2004) enrolled 126 healthy, glucose-tolerant participants (Asian American, African American, white and Mexican American) (age 26.0±6.0 y, BMI=24.7±4.2kg/m^2). A positive correlation was noted between 25(OH)D concentrations with insulin sensitivity

and a negative effect of vitamin D deficiency on ß cell function was seen. Additionally, the researchers demonstrated that subjects with vitamin D deficiency were at higher risk of insulin resistance and the metabolic syndrome.

A study by Need *et al* (2005) included a total of 753 postmenopausal women (aged 63.0±9.4 y, BMI=26.5±5·2 kg/m^2) who were not on any treatment known to affect glucose metabolism. The data indicated that fasting serum glucose increased as 25(OH)D concentrations fell throughout the range of 25(OH)D measured but the greatest increase was observed in those with 25(OH)D below 40 nmol/l.

Forouhi et al (2008) selected randomly 524 European-origin non-diabetic participants (age 40-69 y, BMI=23.3-26.0 kg/m^2) from the population-based Ely Study (Cambridgeshire, UK), which was established in 1990 as a longitudinal cohort study of the etiology and pathogenesis of type 2 diabetes and related metabolic disorders. The result showed inverse associations between baseline 25(OH)D and insulin resistance.

In another study, Liu and others (2009) examined a cross-sectional association between 25(OH)D and markers of the insulin resistant phenotype in 808 Caucasian non-diabetic participants (age 59.6±0.3 y, with a mean of BMI 27.8 kg/m^2). The results recognized an inverse association between 25(OH)D and insulin resistance. Moreover, an inverse association has been observed between 25(OH)D and serum insulin in 3,890 U.S. non-diabetic participants (age 40.0±8.7 y, BMI=26.7±5.3 kg/m^2) (Cheng et al. 2010).

Table 5.1 Results of different studies of vitamin D status and obesity

Reference	**Study population**	**Country**	**Age**(y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**%BF**	**Comments**
Bell et al. (1985)	**White subjects** 14 Non-obese 12 Obese	**USA** S. Carolina	**Range** 20-35	50.0±5.0 20.0±2.5	**Average BW** 68±2 **to** 106±6	- -	Mean 25(OH)D was lower in the obese than in the non-obese subjects
Liel et al. (1988)	**White subjects** 13 Non-obese 13 Obese	**USA** S. Carolina	**Range** 20-35	40.0±5.0 27.5±2.5	- -	-	25(OH)D was significantly lower in the obese compared with the non-obese subjects
Buffington et al. (1993)	**Females** 37 subjects 23 subjects	**USA** Tennessee	36.5±1.8 34.9±2.2	Deficient Non-deficient	50.8±1.8 42.3±1.1	- -	Significant and negative correlation between 25(OH)D and body mass
Need et al. (1993)	**Postmenopausal** 433 subjects	**Finland** Helsinki	60.4±0.44	63.4 **Range** 10-196	25.4 **Range** 15.7-43.1	-	Negative relation between 25(OH)D and BMI
Wortsman et al. (2000)	**White subjects** 13 control 13 obese	**USA** Philadelphia	 34.0±3.0 37.0±2.0	 38.3±5.5 17.4±3.6	 22.2±0.04 38.0±1.7	 -	Inverse correlation between BMI and 25(OH)D $_3$ in subjects received UV-B irradiation.
	11 control 11 obese		36.0±4.0 39.0±3.0	5.3±0.2 3.5±1.5	21.4±0.6 35.7±1.8	 -	Inverse correlation between BMI and 25(OH)D_2 in subjects received pharmacologic dose of vitamin D_2 orally.

Table 5.1 Results of different studies of vitamin D status and obesity

Reference	**Study population**	**Country**	**Age** (y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**%BF**	**Comments**
Arunabh et al. (2003)	**Females** 171 black 239 white	**USA** New York	47.6±14.8	54.2±34.7	23.9±2.9	36.2±7.4	Inverse relation between 25(OH)D and %BF
Parikh et al. (2004)	148 Non-obese 154 obese	**USA** Metropolitan Washington	36.6±11.4 37.6±9.4	77.5±36.0 58.7±30.5	25.66±3 37.44±6	- -	Serum 25(OH)D concentrations was negatively correlated with BMI
Looker (2005)	**Females** Non-Hispanic White 788 970 886 923 Non-Hispanic Black 925 909 458 183	**USA** Maryland	 12-29 30-49 50-69 70+ 12-29 30-49 50-69 70+	 88.57 77.47 68.25 65.95 45.62 42.8 49.97 48.0	 - - 	 33.9 37.4 	The negative relationship between 25(OH)D and % BF was noticeably stronger in whites than in blacks of the same age
Snijder et al. (2005)	237 males 216 females	**Netherlands** Amsterdam	≥65	40.1±17.7 to 59.4±23.9	22.4±2.2 to 32±3.9	19.2±4.1 to 51.4±2.6	Total body fat, BMI and WC were associated negatively with lower 25(OH)D

Table 5.1 Results of different studies of vitamin D status and obesity

Reference	**Study population**	**Country**	**Age**(y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**%BF**	**Comments**
Bischof et al. (2006)	483 Subjects	**Austria** Vienna	48.2±16	52.75±26.75	32.7±8.2	-	Negative relationship between 25(OH)D and BMI
Hahn et al. (2006)	**PCOS*** **Caucasian**	**Germany** Essen					
	32 lean		29.0±5.0	53.2±29.5	22.2±1.7	-	Low 25(OH)D were associated with obesity
	18 overweight		28.6±1.5	48.0±3.0	26.6±1.5	-	
	70 obese		29.1±7.1	37.5±26.2	37.1±4.9	-	
Bolland et al. (2007)	378 males	**New-Zealand** Auckland	57.0±11.0	85±31	26.4±3.4	24±7	levels of fat mass affect peak 25(OH)D
	1606 postmenopausal		74.0±4.0	51±19	26.4±4.3	41±8	
Hyppönen and Power (2007)	7437 Subjects	**UK** South, Midlands & Wales, North, and Scotland	45.0	42.8 36.7	Non-obese obese	-	Obese participants were at the highest risk of hypovitaminosis D.
Aasheim et al. (2008)	**Males**	**Norway**					
	28 control	Southeast	39.0±11.0	59±20	25±3	-	Low concentrations of serum 25(OH)D were prevalent in morbidly obese
	34 morbidly obese		41.0±10.0	34±15	45±7	-	
	Females						
	30 control		39.0±11.0	54±22	23±3	-	
	76 morbidly obese		41.0±11.0	40±16	45±7	-	

* Polycystic ovary syndrome

Table 5.1 Results of different studies of vitamin D status and obesity

Reference	**Study population**	**Country**	**Age**(y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**%BF**	**Comments**
Forouhi et al. (2008)	**Non-diabetic** 214 males 310 females	**UK** Cambridgeshire	52.9 ±7.7	64.7± 27.3 57.1±23.4	- -	- -	An inverse association between 25(OH)D and fasting insulin as well as with waist circumference also
Konradsen et al. (2008)	**Males& Females** 53 /245 99 /493 130/528 72 /323 56 /188	**Norway** Oslo	43.5±18.1 47.0±15.1 48.3±14.2 47.6±13.5 45.5±12.9	83±24.8 76.4±24.9 73.3±28.4 65.9±22.0 63.9±33.6	<25 25-29.9 30-34.9 35-39.9 ≥40	- - - - -	An inverse association between 25(OH)D and BMI
Macdonald et al. (2008)	**Postmenopausal Caucasian** 3113	**UK** Aberdeen	54.8±2.3	< 70 ≥ 70	26.8±4.9 26.1±4.3	-	low 25(OH)D concentrations are associated with obesity
McGill et al. (2008)	250 subjects	**New- Zealand** Auckland	47.6±11.6	62.2±22.7	35.4±5.2	-	An inverse association between 25(OH)D and BMI and waist circumference

Table 5.1 Results of different studies of vitamin D status and obesity

Reference	**Study population**	**Country**	**Age**(y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**%BF**	**Comments**
McKinney et al. (2008)	255 Non-Hispanic White	**USA** Texas	24.3±4.9	71.8±30.2	26.26±6.2	35.3±8.1	Serum 25(OH)D concentrations were negatively associated with % BF
	260 Non-Hispanic black			37.6±17.6	30.27±7.8	35.8±8.5	
	285 Hispanic			47.9±19	28.16±6.3	37.2±6.8	
Kremer et al. (2009)	**Females**	**USA**					
	53 Hispanic	California	19.6±1.4	78.0±36.5	<25	-	Significant negative correlations between 25(OH)D and BMI
				58.2±23.2	≥25		
	37 Caucasian		19.1±1.6	91.5±32.25	<25	-	
				74.2-5±23.2	≥25		
Lagunova et al. (2009)	**Males**	**Norway**					
	42	Oslo	44.8±20.01	81.6±3.11	23.3±1.2	-	Negative correlation between 25(OH)D and BMI
	96		45.2±17.32	75.8±2.74	27.7±1.4		
	129		47.7±15.84	68.7±2.03	32.1±1.3		
	70		46.3±14.58	63.1±2.67	37.2±1.4		
	52		44.1±13.75	53.0±2.8	44.8±4.2		
	Females						
	31		38.8±16.54	78.2±3.99	18.7±1.0	-	
	206		44.2±17.76	84.9±1.81	23.1±1.1		
	482		47.2±14.72	76.5±1.12	27.6±1.4		
	529		48.3±13.7	73.2±1.11	32.3±1.4		
	317		47.7±13.23	66.6±1.23	37.1±1.3		
	179		46.1±12.76	64.9±1.91	44.4±4.5		

Table 5.1 Results of different studies of vitamin D status and obesity

Reference	**Study population**	**Country**	**Age**(y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**%BF**	**Comments**
				Range		**% FM**	
Moschonis et al. (2009)	**Postmenopausal**	**Greece**	59.8±4.5	77.5-118.7	28.3±3.9	42.1±5.2	Total BF was strongly associated with 25(OH)D
	37	Athens	59.7±4.9	52.7-77.5	29.4±6.4	44.0±3.6	
	38		62.0±3.7	25.2-52.7	29.5±4.1	44.9±5.5	
	37						
Pitroda et al. (2009)	103 (16.5% females)	**USA** Boston	71.5±5.3	79.9±3.7	24.8±2.8	24.0±4.5	Serum 25(OH)D concentrations were only marginally correlated with %BF
	102 (50.0% females)		71.0±4.3	81.2±3.3	26.2±3.5	33.8±2.3	
	102 (97.1% females)		70.4±3.9	68.5±4.1	29.2±4.1	43.2±3.7	
Rodriguez-Rodriguez et al. (2009)	**Females**	**Spain**	27.8±4.6	<90	28.6±3.2	-	A BMI of<27.7 was associated with 25(OH)D of ≥90
	66 White Spanish	Madrid		≥90	26.0±1.3		
Winters et al. (2009)	52 Whites	**USA** Louisville	30.4±7.6	38.5±33.75	29.8±7.9	-	BMI was associated with 25(OH)D after adjustment for age in white but not in African American
	36 African American		30.0±7.1	23.75±19.5	29.3±9.2		

Table 5.1 Results of different studies of vitamin D status and obesity

Reference	**Study population**	**Country**	**Age**(y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**%BF**	**Comments**
Yildizhan et al. (2009)	**PCOS***	**Turkey**	26.6±3.6	73±20.2	22.1±1.8	-	Low serum concentrations of 25(OH)D were associated with higher BMI values
	43 non-obese	Van	25.5±3.9	31.7±9.2	32.8±5.4	-	
	57 obese						
Young et al. (2009)	**Males**	**USA**				-	Serum 25(OH)D concentrations were inversely associated with BMI
	380 Hispanics	Texas	38.4±13.4	44.7±16.7	27.9±5.1		
	191 African-American	Los Angeles	42.1±14.3	30.2±14.0	28.4±5.2	-	
	Females						
	537 Hispanics		39.9±12.6	39.2±19.2	28.6±6.3	-	
	248 African-American		39.5±12.7	25.5±13	30.0±7.4	-	
Brock et al. (2010a)	**1**357 males	**USA**	64.0±5.0	3% had <25	27.0±4.0	-	An association between low 25(OH) D with high BMI
	1264 females		63.0±5.0	29% had <50 79% had <80	27.0±6.0	-	

* Polycystic ovary syndrome

Table 5.1 Results of different studies of vitamin D status and obesity

Reference	**Study population**	**Country**	**Age**(y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**%BF**	**Comments**
Cheng et al. (2010)	3890 subjects	**USA** Massachusetts	40.0±8.7	93±46.25	26.7±5.3	-	An inverse association between 25(OH)D and BMI as well as waist circumference also and positive association between 25(OH)D and physical activity levels as well as vitamin D intake also
Muscogiuri et al. (2010)	18 (56% females)	**Italy** Rome	42.3±12.6	63.6±11.0	26.6±2.7	-	An inverse association between 25(OH)D and BMI
	21 (52% females)		39.6±12.1	30.1±9.1	31.7±6.0	-	
Tzotzas et al. (2010)	**Females**	**Greece**	40.6±11.4	59.5±21.75	22.9±1.5	-	Serum 25(OH)D concentrations were low in obese and correlated inversely with BMI as well as waist circumference also
	25 control	Thessaloniki		42.5±15	36.7±4.9	-	
	44 obese						
Farrell et al. (2011)	**Males**	**USA**	48.4±10.0	<50	28.9±4.6	23.7±5.1	Serum 25(OH)D concentrations were negatively associated with different measures of adiposity
	438	Texas	48.9±9.9	50-75	28.3±4.2	22.9±5.4	
	786		50.5±9.8	>75	27.3±3.6	21.6±5.3	
	1093						

Table 5.1 Results of different studies of vitamin D status and obesity

Reference	**Study population**	**Country**	**Age**(y) Mean ±SD	**25(OH)D** nmol/l Mean ±SD	**BMI** (kg/m^2) Mean ±SD	**%BF**	**Comments**
Mason et al. (2011)	**Postmenopausal** 87 control 117 Exercise trial 118 Diet trial 116 Exercise& Diet trails	**USA** Seattle	 57.4±4.4 58.1±5.0 58.1±5.9 58.0±4.4	**Baseline** 51.2 49.5 50.7 55.0 **After 12 months** **Control** 59.5 **Weight loss <5, 5-9.9, 10-14.9 and ≥15% 25(OH)D=** 52, 57, 68 and 73.7, respectively.	**Baseline** 30.7±3.9 30.7±3.7 31.1±3.9 31.0±4.3	**Baseline** 47.3±4.4 47.3±4.1 47.0±4.3 47.4±4.5 -	Increase 25(OH)D was associated with a great degree of weight loss
Forsythe et al. (2012)	118 subjects 109 elderly	**Northern Ireland** Coleraine **Republic of Ireland** Cork	20-40 ≥64	-	-		An inverse association between 25(OH)D and BMI as well as waist circumference also
Johnson et al. (2012)	**Morbidly obese** 690 males 1336 females	**Norway**	45.0 ±12.1 42.2 ±12.2	50.0 ±22.0 53.6 ±22.4	44.6±6.0 44.3 ±5.9	- -	Males seeking weight loss treatment have significantly higher odds of vitamin D deficiency than females

5.2 Aims

Sunlight has long been the most important source of vitamin D. However, individuals living in the UK have an increased risk of low 25(OH)D caused by a lack of sunlight exposure; particularly during winter (Hyppönen and Power 2007).

Diets rich in high energy foods together with sedentary behaviours such as television viewing and computer games has increased the risk of obesity in Great Britain (Skidmore and Yarnell 2004), and it is known that obesity is a risk factor of low serum 25(OH)D concentrations. Findings in chapters 3 and 4 indicated that Middle Eastern people in the UK may be at risk of vitamin D deficiency from low vitamin D intake from the diet or supplements and their dress style which limits their exposure to sun. Furthermore, the findings of chapter 3 and 4 indicated a high prevalence of overweight /obesity. Thus, the aims of this research were to:

- Investigate the effect of seasonal variation in Middle Eastern and Caucasian people in the UK over a 3 month period of reduced daily sunlight (October-January) on serum 25(OH)D concentrations.
- Present the relationship between vitamin D status and body fatness
- Observe the effect of moderate loss of body fat induced by diet and increased physical activity on serum 25(OH)D concentrations and insulin levels.

5.3 Objectives

The objectives of this study were to:

- Compare vitamin D status between Middle Eastern and Caucasian individuals by measuring 25(OH)D over 3 months (October-January).

- Investigate the effect of body fat loss on vitamin D status by measuring and recording the percentage of body fat over the following 3 months (January-April) together with serum vitamin D.
- Measure serum insulin over the 6 month period (October-April).
- Provide the participants with individual advice and recommendations for a healthy diet to reduce daily energy intake and increase physical activity depending on their usual habits to encourage weight loss over a 3 month period.
- Estimate consumption of vitamin D and energy and other nutrients using food diaries.
- Estimate physical activity individual levels via a questionnaire (October-January- April).

5.4 Ethical Approval

The Human Ethics committee of the Faculty of Science /University of Plymouth approved the study protocol. Each participant was provided with a consent form. The consent form stated that each subject has the right to withdraw their information and data collected from the study at any time; and this was verbally reinforced (Appendix 5).

Additionally a subject information leaflet was designed, printed and distributed for individuals at the campus. The leaflet included the title of the study, the contact information of the researcher, supervisor and secretary of the Faculty of Science and Technology Human Ethics Committee. In addition, it contained an introduction about vitamin D along with questions and their answers about the participation in the study; such as: who has approved the project, what would be involved in taking part, will take part be of any benefit to the participant, are there disadvantages to taking part, etc. (Appendix 6).

5.5 Subjects and recruitment

Potential volunteers for the study were recruited on the university campus using posters, via emails and an advertisement on the university intranet.

5.5.1 Advertising the study

- **Poster**

A poster for subjects' recruitment was designed with a brief statement about the role of vitamin D, the link between overweight and vitamin deficiency and why Middle Eastern individuals and people living in the UK are especially at risk. Moreover, the poster contained the objectives of the study with researchers' emails for further information. Several copies of the poster were displayed in different locations inside the campus such as: the nursery, the gym and the canteens (Appendix 7).

- **Email/electronic- advertisements**

Two advertisements were distributed via the University intranet to catch individual's attention and call for volunteers for the study; the first advert was submitted as an email via the "International student advisory service" and the second advert was put on the "staff announcements" for the UOP intranet.

5.5.2 Enrolment of subjects

The enrolment process was started by reading and signing the consent form, which indicating a willingness to volunteer for the research study.

5.5.2.1 Inclusion criteria for potential participants

- Middle Eastern and Caucasian subjects - males and females
- Aged at least 18 years
- High percentage of body fat (> 20% for males and > 30% for females) (Grodner et al. 2012)
- Wishing to lose weight

5.5.2.2 Exclusion criteria for potential participants

- Low and normal percentage of body fat (< 20% for males and < 30% for females) or BMI<25 kg/m^2
- Intake of all supplements containing vitamin D
- Dieted in last 3 months
- Pregnant or lactating
- Subjects with prescription medications (unless known not to effect serum 25(OH)D concentrations and the percentage of body fat).
- Have a pace- maker (individuals with a pacemaker were excluded from the study because BIA affects the functioning of pacemakers).
- Upon completion of the consent form, each subject which met the inclusion criteria was shown how to complete a 3 day food diary and they were asked to return the completed food diary form to Nutrition & Food Lab.

5.6 Calculation of sample size required

A power calculation was used (University of British Colombia 2011) to determine the sample size required for fat mass reduction advice, using a mean weight loss of 10.4 kg and standard deviation (SD) of 12.5 kg, taken from another study assessing weight loss, serum 25(OH)D concentrations and insulin resistance (Tzotzas et al. 2010). In order to detect a mean differences in 25(OH)D after 12 week body fat reduction programme 23 participants were required to detect 80% power at the 0.05 significant level. In order to allow for a 25% drop out rate 29 participants would be recruited in the study.

5.7 Experimental protocol

Table 5.2 summarized the time line of study protocol, which consisted of two phases:

5.7.1 Phase 1-observational (week 1-12 or October-January)

In order to observe the effect of season on vitamin D status, this part of the study ran between October and January. For the duration of this time all participants were encouraged to follow their usual eating habits and maintain their usual physical activity (Table 5.2). Phase 1 included:

- **Measurements taken at baseline and at 12 weeks**

- Demographic/ screening questionnaire
- Anthropometric measurements (Body weight, BMI, body fat, waist circumference and waist-hip ratio)
- Food diary
- Physical activity questionnaire
- Blood samples for serum 25(OH)D concentrations and serum fasting insulin

5.7.2 Phase 2-intervention (week 12-24 or January - April)

In this part of the study volunteers were advised to lose weight and increase their physical activity to examine the effect on serum 25(OH)D concentrations. Qualified dietitians met the subjects once a month during the weight loss period to give individual dietary advice according to the participant's dietary records. Advice was given to reduce usual energy intake and increase physical activity of each subject to encourage weight loss over a 3 month period. Monthly anthropometric measurements were taken (Body weight, BMI, body fat, waist circumference and waist-hip ratio).

- **Week 24 measurements (final measurements)**

- Demographic /screening questionnaire to measure change in status between baseline (week1) and week 24.
- Anthropometric measurements
- Blood samples for serum 25(OH)D concentrations and serum fasting insulin

Table 5.2 Time line of experimental protocol (seasonal variation and fat loss effect)

Time / Method	**Phase 1:** Seasonal variation			**Phase 2:** Loss of body fat		
	Oct. 2010 Week 1	**Nov. 2010** Week 8	**Jan.2011** Week 12	**Feb. 2011** Week 16	**March 2011** Week 20	**April 2011** Week 24
Demographic Screening questionnaire	✓		✓			✓
Anthropometric measurements	✓		✓	✓ *	✓ *	✓
Food diary	✓		✓			**
Physical activity questionnaire	✓		✓			✓
Blood samples	✓		✓			✓
Individualized dietary advice			✓	✓	✓	✓

* Measurements taken to aid individual advice. Records not presented

** Since food diary incur burden for the participants, the decision was made to stop it at week 24

5.8 Experimental tools

5.8.1 Demographic screening questionnaire

The questionnaire was administered at baseline (week 1) and at the end of the study (week 24). It contained two sections:

- **personal information:** age, gender, ethnic origin, smoking, weight lost during the last three months, following a special diet, outdoor activities with exposure to sunlight, sunscreen use and sun protection factors, dress style, occupation.
- **Medical information:** prescribed or over the counter medications including the contraceptive pill/ hormone replacement therapy (HRT) for women, nutritional supplement (vitamins/minerals), and having a pacemaker (Appendix 8).

5.8.2 Anthropometric measurements

5.8.2.1 Body Mass Index (BMI)

Height (cm) was recorded to the nearest 0.5 centimeter using a stadiometer (Model seca 225, Hamburg, Germany). Body weight (kg) was measured to nearest 0.1 kilogram. Weight status was defined using Body Mass Index (BMI) which is calculated by dividing body weight in kilograms by height in meters squared (kg/m^2).

5.8.2.2 Waist-hip ratio (WHR)

Waist and hip circumferences were measured to the nearest 0.001 millimetre using a stretch-resistant measuring tape (Visser et al. 1997, WHO 2008). Subjects were stood with arms at the sides, feet positioned close together. The tape was held snugly, but not constricting, and parallel to the floor at the level at which the measurement is made (WHO 2008). Waist circumference was assessed at the point of the minimal waist (Ross et al. 2008). Hip

circumference was taken around the widest portion of the buttocks. Waist-hip ratio (WHR) was calculated as waist circumference divided by hip circumference (HC). The WHO cut-off points for waist and hip circumference and for WHR were adopted to define and risk of metabolic complications (WHO 2008) (Table 5.3).

Table 5.3 WHO cut-off points and risk of metabolic complications

Indicator	Cut-off points		Risk of metabolic complications
	Males	Females	
WC	>94 cm	>80 cm	Increased
WC	>102 cm	>88 cm	Substantially increased
WHR	≥0.90 cm	≥0.85 cm	Substantially increased

5.8.2.3 Body composition

Body composition and total body fat was estimated by using a body composition analyser [foot-to foot tanita bioelectrical impedance analysis (BIA)] (Model TBF-300M/TBF-300MA, Birmingham, UK). Subjects were asked to remove their shoes and socks. To ensure normal hydration status for BIA testing, subjects were asked to adhere to the following pre-test requirements (Dixon et al. 2008):

- No eating or drinking for 4 h before testing
- No caffeine or alcohol consumption within 48 hours of the test
- No vigorous exercise within 12 hr of the test
- Empty bladder 30 minutes before test

5.8.3 Recording of physical activity levels

The short version (self-administered) of the international physical activity questionnaire (IPAQ) (www.ipaq.ki.se) was used to obtain comparable estimates of physical activity at week 1, 12 and 24 of the study. The questionnaire asked the subjects about the time that they spent being physically active in the last 7 days for at least 10 minutes at a time. The IPAQ asked about

three specific types of activity; walking, moderate-intensity activities and vigorous-intensity activities (Appendix 9).

IPAQ was structured to provide separate scores on each type of activity. Computation of the total score for this form required summation of the duration (in minutes) and frequency (days) of walking, moderate-intensity activities and vigorous-intensity activities. Both categorical variables (low, moderate and high) and continuous indicators of physical activity would be used to classify the sample. The continuous indicator was presented as median minutes/week (median MET-minutes/week) and it was computed by weighting each type of activity by its energy requirements defined in METs to yield a score in MET-minutes. METs are multiples of the resting metabolic rates, and the MET-minute is computed by multiplying the MET score of an activity by the minutes.

The following values were used for the analysis of IPAQ data: Walking=3.3 METs, Moderate PA=4.0 METs and Vigorous PA=8.0 METs. Using these values, four continuous scores were defined as the equations below:

- Walking MET-minutes/week =3.3 * walking minutes * walking days
- Moderate MET-minutes/week=4.0 * moderate-intensity activity minutes * moderate days.
- Vigorous MET-minutes/week=8.0 * vigorous-intensity activity minutes * vigorous-intensity days.

Total physical activity MET-minutes/week=sum of (Walking + Moderate + Vigorous MET minutes/ week scores) .

Categorical score-three levels of physical activity are used as below:

- **Low**

- No activity is reported or
- Some activity is reported but not enough to meet categories 2 or 3.

- **Moderate**

Any one of the following criteria:

- 3 or more days of vigorous activity of at least 20 minutes per day
- 5 or more days of moderate-intensity activity and/or walking of at least 30 minutes per day
- 5 or more days of any combination of walking, moderate-intensity or vigorous intensity activities achieving a minimum of at least 600 MET-minutes/week.

- **High**

Any one of the following criteria:

- vigorous intensity activity on at least 3 days and accumulating at least 1500 MET-minutes/week
- 7 or more days of any combination of walking, moderate-intensity or vigorous intensity activities accumulating at least 3000 Met-minutes/week.

5.8.4 Dietary data collection and editing

Self-completion food and drink diaries were used to assess food consumption and nutrients intake (Food Standard Agency 2010). Subjects were asked to record details of all food and beverages consumed at home and outside the home at the time of consumption over 3 days. All the subjects were asked not to change what they normally ate and drank and to record their eating as they go, not from memory at the end of the day. They were asked to use written notes on a pad if they forget to take their diary with them. Each diary day covered a 24 hr. period, so the subjects were asked to include any food or drinks that they may have had during the night and remember to include foods and drinks between meals including water. Each subject was provided with an instruction booklet (Appendix 10) and a blank food diary form (Appendix 11). Energy and nutrient estimations were calculated using nutritional analysis software package (Dietplan6 2008). For calculation purposes, Middle Eastern recipes were analysed to ingredient values when dealing with Middle Eastern dishes

containing multiple ingredients. As in Chapter 4 when using Compeat, Dietplan does not contain traditional Middle Eastern dishes and therefore details of recipe ingredients were required from participants. Participants were asked to give details of recipe ingredients in the recipe section of the booklet. The proportion of each ingredient was estimated from the overall portion size eaten and this was manually entered onto Dietplan. The decision was taken to manually enter each recipe ingredient into the individual's profile rather than creating recipes on the databases as quite frequently the recipes differed between individuals and a standard recipe that suited all could not be found. Often people changed the ingredients in their recipes according to the available ingredients in the home and thus it wasn't always the same even if eaten on more than one occasion so finding a standard recipe was difficult and would have introduced error. The decision to change to Diet Plan was because the university purchased this package between the studies in chapters 4 and 5 and the database for Dietplan was more up to date than Compeat.

Underreporting was calculated by dividing reported energy intake by predicted basal metabolic rate (EI/BMR) (FAO/WHO/UNU 2004) (See chapter 4).

5.9 Procedure during phase 2 (week 12- 24)

Participants were seen at week 12, 16, 20 and 24 to receive individualised nutritional counselling by a registered dietitian. Monthly anthropometric measurements and nutritional analysis/consultation were recorded in an individual file for each participant. The files contained the following sheets:

- Data collection sheet (Appendix 12)
- Researcher record sheet (Appendix 13)
- Consultation record sheet / Advice and notes (Appendix 14)

At the start of phase 2 each participant was provided with a copy of the record sheet/Goals and targets (Appendix 15), 5 A DAY poster guide (NHS 2011) (Appendix 16), and “Want to lose weight and keep it off” sheet (BDA 2008) (Appendix 17).

5.10 Laboratory assays

Serum vitamin D and fasting serum insulin samples were prepared according to the procedure outlined by Maunsell and others (2005). Blood samples were collected from the subjects after 12 h overnight fast into serum-separating tubes (SST) (BD Vacutainer Systems) and were left for 20 minutes and then centrifuged at 1200g for 10 minutes. Subjects were allowed to have water and their own medications. Blood samples were taken in a quiet clinical room aside for this purpose by a trained phlebotomist. The serum thus obtained was stored at -20 °C before analysis in small plastic vials labelled with personal information for each subject (date of birth, initials, gender, study code and date of collection) (Maunsell et al. 2005).

Each subject was given an alpha-numerical code and this list was kept separate from the study notes and electronic data, so enhancing confidentiality.

In week 24 after all the samples had been collected they were sent to the Derriford Combined Laboratory, Derriford Hospital, Plymouth, UK. Total serum 25(OH)D concentrations were measured using isotope-dilution liquid chromatography-tandem mass spectrometry (ID-LC-MS/MS) (Maunsell et al. 2005). Fasting insulin levels were determined using a solid-phase enzyme-labeled chemiluminescent immunomertic assay kit (IMMULITE 2000 Insulin; The Quality System of Siemens Healthcare Diagnostics Products Ltd, ISO, USA), according to the manufacture instructions by Derriford Hospital. Vitamin D status has been categorized to three groups as follows: vitamin D deficiency,

25(OH)D concentrations<25 nmol/l, vitamin D insufficiency, 25(OH)D concentrations <25 and >50 nmol/l, and vitamin D sufficiency, 25(OH)D concentrations <50 and >300 nmol/l. The normal range for insulin levels was 0-16 mu/l.

5.11 Statistical analysis

Statistical analysis was carried out using Excel (2010) and Minitab (version 16.0, Ltd, Coventry) programmes. Data are expressed as mean ± SD and range. Chi-square statistics were used to compare and to find differences in physical activity levels between ethnic groups. The seasonal effect on mean fasting serum 25(OH)D concentrations and fasting serum insulin in summer (October) and winter (January) within gender and ethnic groups was assessed using one-way ANOVA. Differences between ethnic groups and characteristics before and after 16 week weight loss were estimated using paired T-test. Relationships between variables were evaluated with Pearson correlation coefficients. *P* value <0.05 was considered statistically significant.

5.12 Results

5.12.1 Phase 1 - observational (week 1- 12) October - January

5.12.1.1 Baseline characteristics (week 1)

The advert on the University portal was an effective tool and successful in attracting potential subjects during the first week of operation. More than 75 emails and requests were received from individuals who were wishing to participate in the study. Some declined the invitation to participate mainly due to reluctance to give blood samples. Forty seven subjects were screened based on study-specific criteria. Six were excluded from the study as they did not match the inclusion criteria. Forty one volunteers were included and enrolled into the study.

A total of 41 volunteers, fourteen Middle Eastern and 27 Caucasians, were recruited to the study lasting 7 months (between October 2010 and March/April 2011). All Middle Eastern subjects were of Iraqi decent, and all the subjects lived in Plymouth, which is located in South West England, at latitude 50°N.

During the research study, 2 Middle Eastern, and 3 Caucasian subjects left the study for different reasons: 1 left the UK, 1 left due to language problems, 2 developed health problems and 1 gave no reason.

A total of 36 overweight and obese subjects completed the study. The mean age was 43 ±9.8 y (range: 28-62 y). Overall, 27(75%) of subjects were females and 12 (33.3%) were Middle Eastern. A total of 50% of Middle Eastern females reported sunlight avoidance using clothing. Males accounted for 6 (50%) of the Middle Eastern group, and 3 (13%) of Caucasians. All baseline descriptive characteristics of the study sample according to ethnicity are presented in table 5.4.

Table 5.4 Baseline characteristics of the study sample (week 1)

Variables	Middle East (n=12)	Caucasian (n=24)	*P* value
Gender	6 (50%) males	3 (13%) males	0.039
Age (y) Mean ±SD **range**	38.2±6.1 30-49	45.4±10.6 28-62	0.014
Current smoker [n (%)]	1 (11.1)	5 (18.5)	0.522
Sunscreen use [n (%)]	2 (16.6)	18 (75)	0.0001
Sun protection factor range	15-50	15-60	-

5.12.1.2 Anthropometric information (week 1)

Table 5.5 shows the comparison of anthropometric variables, and body composition between Middle East and Caucasian subjects according to gender. No significant differences were found in height, BMI, basal metabolic rate, waist and hip circumferences, waist-hip ratio, percentage of body fat, total body fat mass, fat free mass and total body water between Middle Eastern and Caucasian males.

The mean age for Middle Eastern females was 36.3 y, significantly younger than the mean age for Caucasians females of 46.2 y.

The mean height for Middle Eastern females was 157.3 cm, significantly lower than the mean height for Caucasians females of 162.4 cm.

There were no significant differences in mean body weight, BMI, basal metabolic rate, waist and hip circumferences, waist-hip ratio, percentage of body fat, total body fat mass, fat free mass and total body water by ethnicity for females.

Table 5.5 Baseline anthropometric information of the sample according to ethnicity (week 1)

Variables	Males			Females		
	Middle East (n=6)	**Caucasian (n=3)***	***P* value**	**Middle East (n=6)**	**Caucasian (n=21)**	***P* value**
Age (y)	40.0±7.9	40.0 (31-46)	1.00	36.3±3.6	46.2±10.9	0.002
Height (cm)	175.3±6.3	180.3 (174-187)	0.351	157.3±3.7	162.4±4.8	0.019
Body weight (kg)	89.1±7.3	101.5 (92.4-111.6)	0.146	81.4±15.2	80.7±14.8	0.917
BMI (kg/m^2)	29.0±1.3	31.5 (28.7-36.9)	0.461	32.8±5.4	30.4±4.6	0.356
Basal metabolic rate (Kcal)	1907±152	2096 (1926-2186)	0.146	1558±150	1514±149	0.543
Waist circumference (cm)	101±8.4	108.3 (98.0-122.0)	0.452	92.5±13.5	89.5±11.1	0.63
Hip circumference (cm)	106.3±3.2	112.7 (106.0-114.0)	0.215	112.3±11.9	110.7±9.0	0.78
Waist-hip ratio (cm)	0.95±0.07	0.96 (0.88-1.03)	0.846	0.82±0.07	0.82±0.07	0.814
Body fat (%)	25.4±3.3	28.5 (24.9-33.2)	0.36	40.2±5.9	39.6±5.06	0.829
Fat mass (kg)	22.7±3.2	29.1 (25.0-37.1)	0.267	33.4±11.4	32.5±9.8	0.862
Fat free mass (kg)	66.5±6.1	72.4 (67.2-75.4)	0.163	48.0±4.0	48.2±5.8	0.937
Total body water (kg)	48.7±4.5	53.0 (49.2-55.2)	0.165	35.2±2.9	35.3±4.3	0.941

Data are expressed as mean ±SD
*The range has been used instead of SD due to small sample size

5.12.1.3 Baseline physical activity levels (week 1)

Figure 5.1 shows self-reported physical activity levels of the study sample according to ethnicity (week 1). The chi-square with two degree of freedom (x^2 2) showed no significant differences in physical activity levels between the two ethnic groups (*P*=0.967). Among the Middle Eastern group 33.3% of the subjects were highly active, 25.0% and 41.6% were of moderate and low activity, respectively; while in Caucasian sample 12.5%, 58.3% and 29.1% of the subjects were considered as high, moderate and low activity, respectively.

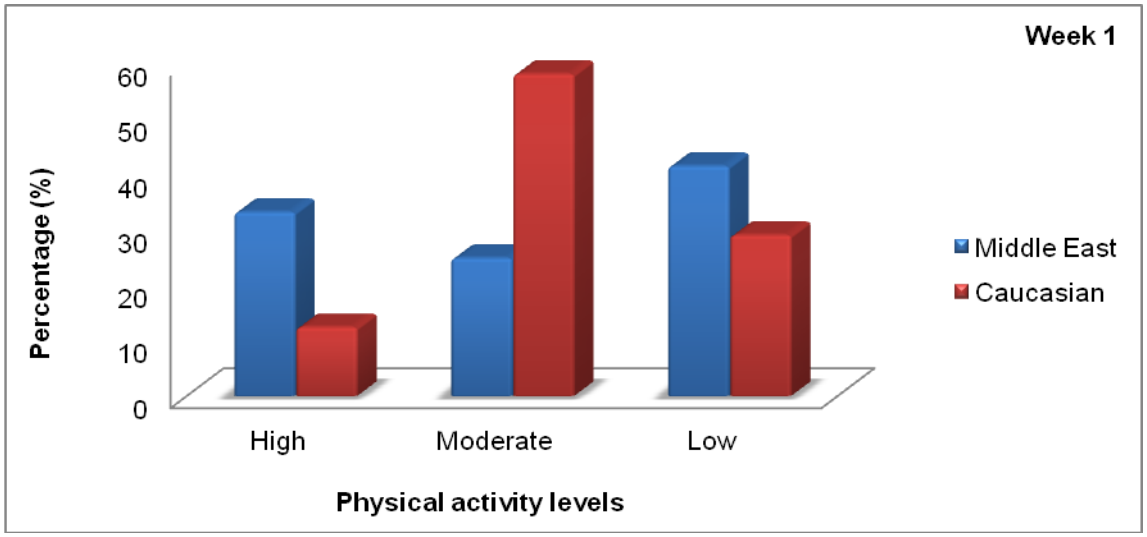

Figure 5. 1 Physical activity levels of the study sample according to ethnicity

5.12.1.4 Baseline dietary intake (week 1)

Figure 5.2 illustrates the mean daily intake of vitamin D (µg), calculated from the 3 days food diary data at baseline of the seasonal variation study (week 1) in the two ethnic groups. No significant difference was found between mean daily intake of vitamin D (µg) of Middle Eastern (2.4±1.2) and Caucasians (2.8±2.1).

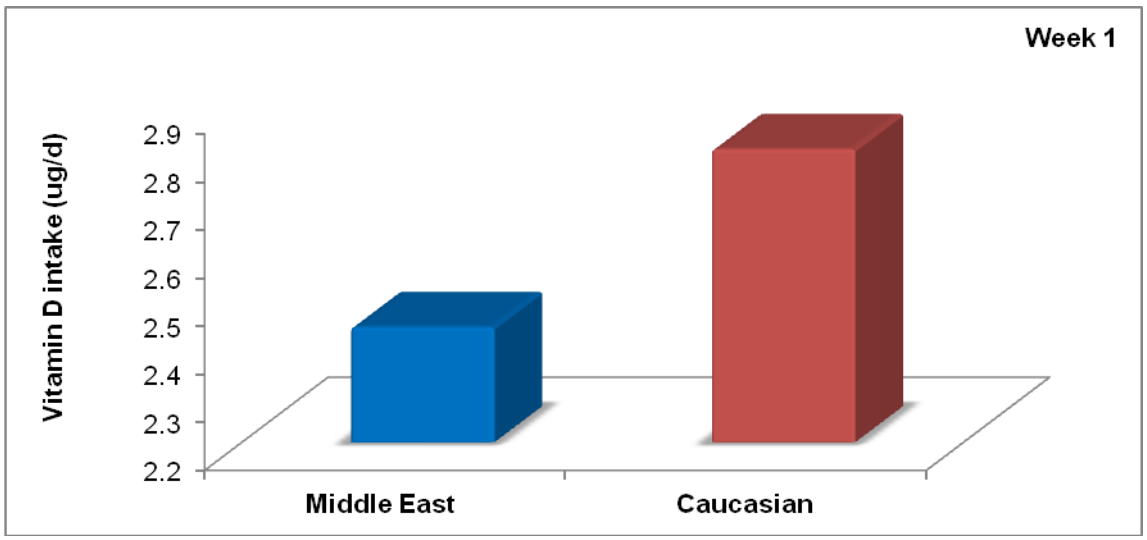

Figure 5. 2 Vitamin D intake of the study sample according to ethnicity

No significant differences in mean daily intake of energy, protein, carbohydrates, fat and vitamin D were found between Middle Eastern and Caucasians according to gender at baseline (week 1) of the study. The mean daily intake of energy was higher than the recommended in both ethnic groups. However, a total of 16.7% of Middle Eastern subjects (16.7% males and 16.7% females) and 16.7% of the Caucasian subjects (about 19% of the females group) were under-reporters according to Goldberg *et al* (1991) equation cut off limits to assess underreporting (EI/BMR <1.14) (Calculated as in chapter 4). Protein and carbohydrate intakes in Middle Eastern males and fat intake in Caucasian males were higher than the recommended. Middle Eastern males recorded higher protein and carbohydrates intakes, and the Caucasian males recorded higher fat intake than the recommended. The mean daily intake of vitamin D was below the Reference Nutrient Intakes (RNIs) (FAO and WHO 2002) in all groups (Table 5.6).

Table 5.6 Energy and dietary intake* of Middle Eastern and Caucasians groups compared to DRV ** according to gender (week1)

Energy and Nutrients	Males				Females			
	Middle East (n=6)	Caucasians*** (n=3)	DRV	*P* value	Middle East (n=6)	Caucasians (n=21)	DRV	*P* value
Energy intake (kcal/d)	2753±625	2958 (2629-3357)	2605****	0.561	2473±696	2200±503	2079****	0.405
%Energy compared to recommendations	105.7	113.5			118.9	105.8		
Protein (g/d)	134.5±62.2	99.3 (78.5-116.2)	55.5	0.251	136.9±53.8	89.5±22.0	45.0	0.089
% Energy as protein	18.9	14.5	15%		21.4	16.4	15%	
Carbohydrates (g/d)	351.8±80.6	293.5 (266.3-333.1)	-	0.182	311.1±85.2	228.6±65.7	-	0.071
% Energy as carbohydrates	49.5	42.9	50%		48.7	41.9	50%	
Fat (g/d)	99.8±26.0	129.2 (112.0-148.0)	-	0.106	84.5±22.9	101.0±31.9	-	0.187
% Energy as fat	31.6	42.5	35%		29.8	41.7	35%	
Vitamin D intake (µg/d)	2.5±1.3	3.8 (1.5-2.8)	5*****	0.532	2.4±1.1	2.7±2.0	5****	0.685

*Data are expressed as mean ±SD
**Dietary Reference Values for Food, Energy and Nutrients for the UK (Department of Health 1991)
***The range has been used instead of SD due to small sample size
****(SACN 2011)
***** RNIs (FAO and WHO 2002)

5.12.2 Serum 25(OH)D and insulin levels and seasonal variation effect

Of the 36 subjects who participated in this study at baseline (week 1, October) 14 (39%) had insufficient vitamin D levels (serum 25(OH)D 25-50 nmol/l), and among them 9 (25%) suffered from vitamin D deficiency (<25 nmol/l) (as categorised by Derriford hospital clinical chemistry department).

No significant differences were found in insulin levels according to ethnicity and seasonal variation. Moreover, no significant differences were found between summer and winter in insulin levels in both ethnic groups (Figure 5.3).

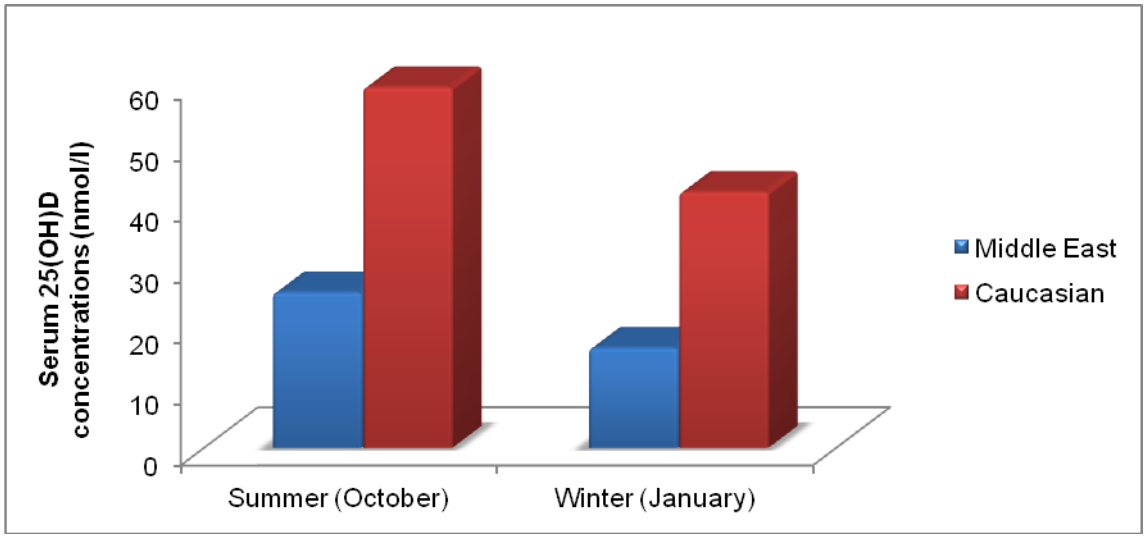

Figure 5. 3 Seasonal variations of 25(OH)D according to ethnicity

Table 5.7 shows the serum 25(OH)D concentrations between two ethnic groups. The mean 25(OH)D among Caucasians were significantly higher than among the Middle Eastern group in summer and winter (*P*=0.0001). Among Middle Eastern and Caucasian subjects, mean 25(OH)D in summer were (25.5±11.5 nmol/l) and (59.2±24.5 nmol/l); respectively. In addition, the percentages of prevalence of vitamin D insufficiency and deficiency in summer and winter were higher in Middle East than Caucasian subjects.

Table 5.7 Comparison of 25(OH)D and SFI by ethnicity according to season

	Middle East (n=12)	**Caucasian (n=24)**	***P* value**
Summer (October)			
25(OH)D (nmol/l) Mean ± SD	25.5±11.5	59.2±24.5	0.0001
Vitamin D insufficiency * (25-50 nmol/l) [n (%)]	4 (33.3)	10 (41.6)	-
Vitamin D deficiency* (<25 nmol/l) [n (%)]	8 (66.6)	1 (4.1)	-
Serum Fasting Insulin ** (mu/l) Mean ± SD	9.0±8.8	7.3±5.1	0.551
Winter (January)			
25(OH)D (nmol/l) Mean ± SD	16.4±5.4	42.0±20.5	0.0001
Vitamin D insufficiency* (25-50 nmol/l) [n (%)]	1 (8.3)	12 (50)	-
Vitamin D deficiency* (<25 nmol/l) [n (%)]	11 (91.6)	5 (20.8)	-
Serum Fasting Insulin** (mu/l) Mean ± SD	8.0±8.0	7.8±5.2	0.948

*According to Derriford Hospital, Plymouth, UK

** According to Derriford Hospital, Plymouth, UK; the normal levels of SFI between 0-16 (mu/L)

5.12.3 Phase 2-intervention (week 12-24) January-April

5.12.3.1 Physical activity levels at the start of phase 2 (week 12)

Figure 5.4 shows self-reported physical activity levels of the study sample at the baseline (week 1) and at the start of phase 2 (week 12). No significant differences in physical activity levels between baseline of phase 1 and week 12 of the total sample. At the baseline, 19.4% of the sample were highly active, 47.2% and 33.3% were of moderate and low activity, respectively. At week 12, a total of 16.7% were highly active, 47.2% and 36.1% were of moderate and low activity, respectively.

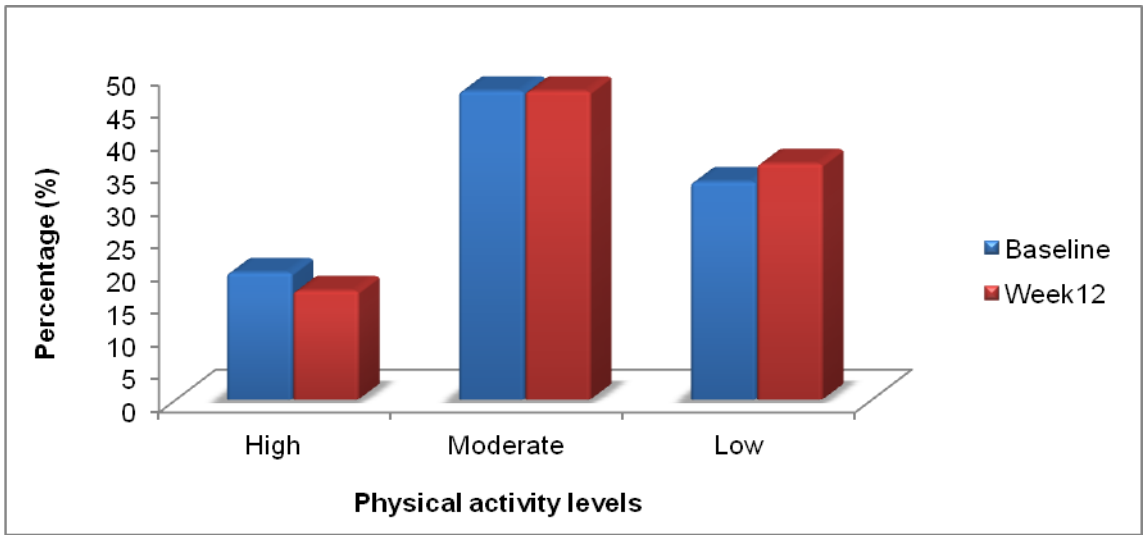

Figure 5. 4 Physical activity levels of the study sample at baseline and week 12

5.12.3.2 Dietary intake of the total sample at week 12 (n=36)

The mean values and standard deviations for recorded daily intakes of energy and other nutrients, at the start of phase 2 (week 12) for the total sample according to gender are presented in table 5.8. Significant differences were observed between males and females in their daily intake of energy, protein, carbohydrates and fat. No significant difference was seen in energy and macronutrient intake between week 1 and week 12 for males and females.

However, no significant difference was between males and females in their daily intake of vitamin D. The mean daily intake of protein was higher the recommended in two gender groups, and the carbohydrate intake in males were slightly higher than the recommended. On the other hand, energy and fat intakes were less than the recommended: 44% of males and 74% of females in the sample reported consuming less than the estimated average requirement for energy, this showing a likely energy deficit in their diet.

Vitamin D intake was less than the recommendation in both males (1.8±1.2 µg/d), and females (2.6±2.1 µg/d) (Table 5.8). No significant differences were found in mean daily intakes of vitamin D (µg) of the total sample between week1 (2.7±1.8 µg/d) and week 12 (2.4±2.0 µg/d).

Table 5.8 Energy and dietary intake of study sample compared to DRV ***** according to gender at the start of phase 2 (week 12)

	Males(n=9)		Females (n=27)		
Energy and Nutrients	**Mean ±SD**	**DRV**	**Mean ±SD**	**DRV**	***P* value between males and females**
Energy intake (kcal/d)	2448±616	2605**	1584±338	2079**	0.003
%Energy compared to recommendation for energy	94.0		76.2		
Protein (g/d)	101.8±26.9	55.5	71.6±20.7	45.0	0.010
% Energy as protein	16.2	15%	17.9	15%	
Carbohydrates (g/d)	318.9±101.2	-	194.8±51.3	-	0.006
% Energy as carbohydrate	50.8	50%	48.6	50%	
Fat (g/d)	92.1±33.3	-	59.6±17.9	-	0.021
% Energy as fat	33.0	35%	33.5	35%	
Vitamin D intake (µg/d)	1.8±1.2	5***	2.6±2.1	5***	0.198

***** Dietary Reference Values for Food, Energy and Nutrients for the UK (Department of Health 1991)
**(SACN 2011)
*** RNIs (FAO and WHO 2002)

5.12.4 Correlation between 25(OH)D and other characteristics of the study sample (week 12)

Table 5.9 shows Pearson correlation coefficient analysis results for the whole study sample (n=36) at the start of phase 2 (week 12). There were significant correlations between serum 25(OH)D concentrations with both waist circumference (r= -0.409, *P*=0.0.013) (Figure 5.5) and waist-hip ratio (r= -0.417, *P*=0.011) (Figure 5.6).

A significant positive correlation were found between 25(OH)D with both dietary intake of vitamin D (r= 0.459, *P*=0.005) (Figure 5.7) and physical activity (r= 0.365, *P*=0.029) (Figure 5.8).

No significant correlations were found between serum 25(OH)D concentrations and both fat mass (Figure 5.9) and serum fasting insulin (Figure 5.10).

Serum fasting insulin correlated positively with age (r= 0.346, *P*=0.039), waist circumference (r= 0.441, *P*=0.007), and waist-hip ratio (r= 0.530, *P*= 0.001). While a marginally significant negative correlation was found between serum insulin and physical activity (r= -328, *P*=0.051) (Table 5.9).

Fat mass correlated negatively with physical activity (r= -0.425, *P*=0.01) (Table 5.9).

Between baseline (week 1) and the start of phase 2 (week 12), the physical activity, dietary intake of vitamin D, weight, body fat and serum insulin were unchanged. Only serum 25(OH)D concentrations changed during this time.

Table 5.9 Correlation coefficient between 25(OH)D and other characteristics of the study sample at week 12 (n=36)

	SFI (mu/l)	**BW** (kg)	**BMI** (kg/m^2)	**WC** (cm)	**HC** (cm))	**WHR** (cm)	**BF** (%)	**FM** (kg)	**FFM** (kg)	**Vit.D intake** (µg/d)	**Physical activity**
25(OH)D (nmol/l)	-0.120 *P*=0.484	-0.262 *P*=0.122	-0.263 *P*=0.121	**-0.409 *P*=0.013**	-0.084 *P*=0.628	**-0.417 *P*=0.011**	0.055 *P*=0.751	-0.109 *P*=0.528	-0.266 *P*=0.117	**0.459 *P*=0.005**	**0.365 *P*=0.029**
SFI (mu/l)		0.221 *P*=0.196	0.132 *P*=0.441	**0.441 *P*=0.007**	-0001 *P*=0.995	**0.530 *P*=0.001**	0.010 *P*=0.953	0.129 *P*=0.454	0.190 *P*=0.266	-0.194 *P*=0.256	**-0.328 *P*=0.051**
BW (kg)			**0.792 *P*=0.0001**	**0.864 *P*=0.0001**	**0.741 *P*=0.0001**	**0.460 *P*=0.005**	0.152 *P*=0.377	**0.692 *P*=0.0001**	**0.766 *P*=0.0001**	0.073 *P*=0.67	-0.190 *P*=0.268
BMI (kg/m^2)				**0.686 *P*=0.0001**	**0.853 *P*=0.001**	0.162 *P*=0.344	**0.587 *P*=0.0001**	**0.903 *P*=0.0001**	0.291 *P*=0.085	0.063 *P*=0.714	**-0.411 *P*=0.013**
WC (cm)					**0.539 *P*=0.001**	**0.773 *P*=0.0001**	0.077 *P*=0.656	**0.557 *P*=0.0001**	**0.699 *P*=0.001**	-0.136 *P*=0.428	-0.242 *P*=0.155
HC (cm)						-0.115 *P*=0.504	**0.599 *P*=0.0001**	**0.869 *P*=0.0001**	0.250 *P*=0.142	0.134 *P*=0.434	**-0.351 *P*=0.036**
WHR (cm)							**-0.366 *P*=0.028**	-0.004 *P*=0.979	**0.640 *P*=0.0001**	-0.258 *P*=0.129	-0.035 *P*=0.839
BF (%)								**0.814 *P*=0.0001**	**-0.515 *P*=0.001**	-0.051 *P*=0.769	**-0.427 *P*=0.009**
FM (kg)									0.066 *P*=0.703	0.066 *P*=0.703	**-0.425 *P*=0.01**
FFM (kg)										0.043 *P*=0.804	0.116 *P*=0.499
Vit.D intake (µg/d)											0.075 *P*=0.663

SFI= Serum fasting insulin
BW=Body weight
BMI=Body mass index
WC=Waist circumference
HC=Hip circumference
WHR=waist-hip ratio
BF=Body fat
FM= Fat mass
FFM=Fat free mass

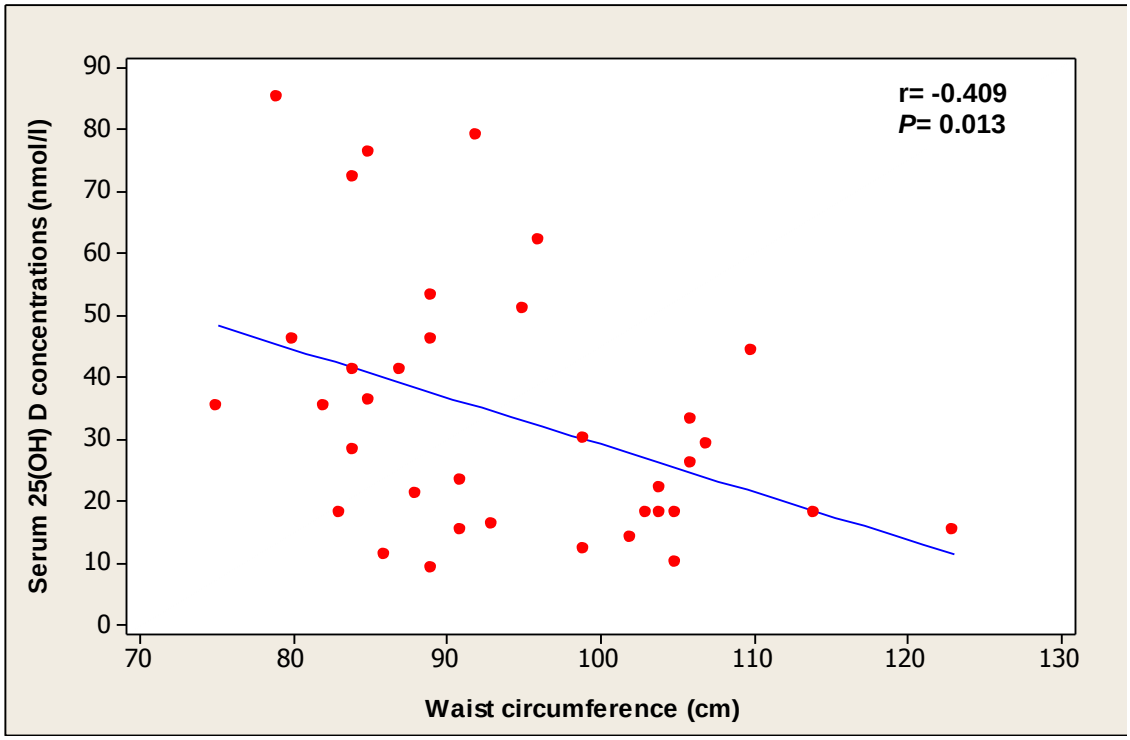

Figure 5. 5 Correlation between serum 25(OH)D concentrations and waist circumference in total sample (phase1-week12)

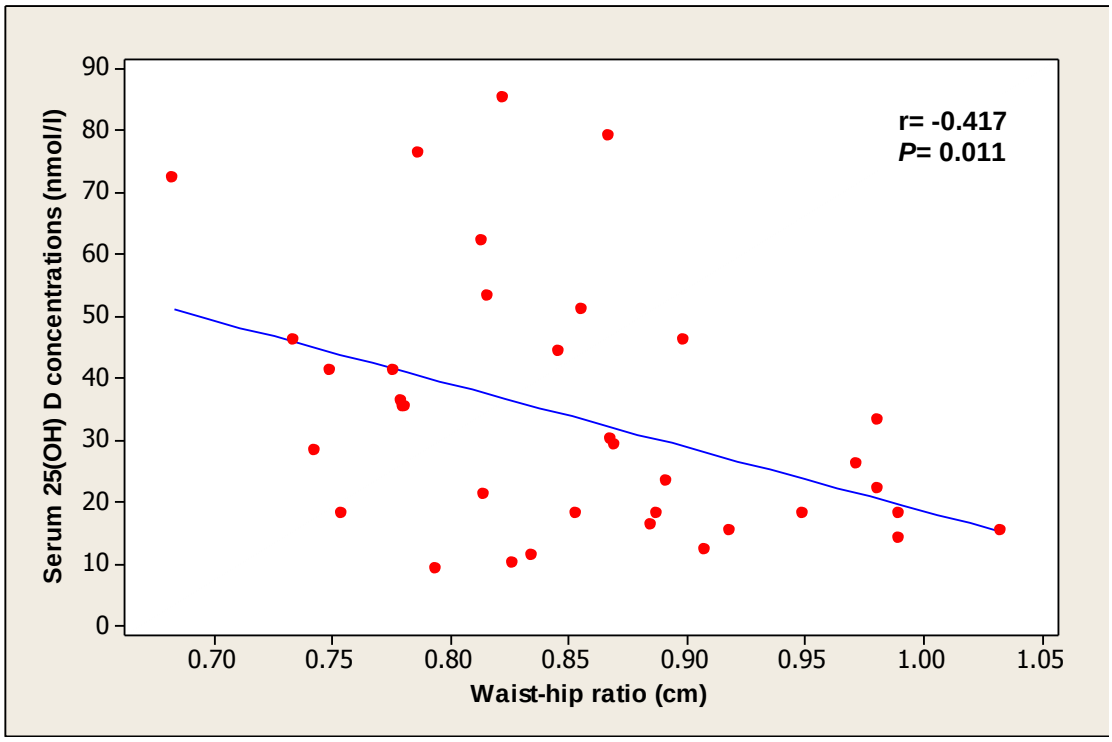

Figure 5. 6 Correlation between serum 25(OH)D concentrations and waist-hip ratio in total sample (phase1-week12)

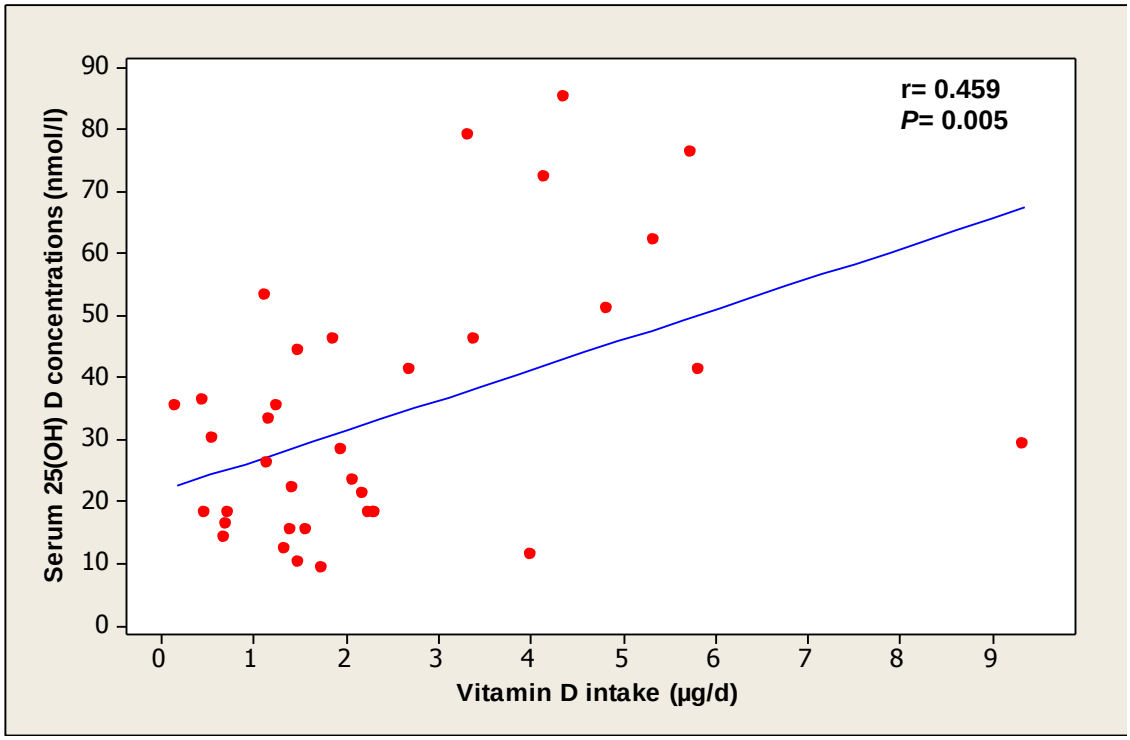

Figure 5. 7 Correlation between serum 25(OH)D concentrations and vitamin D intake in total sample (phase1-week12)

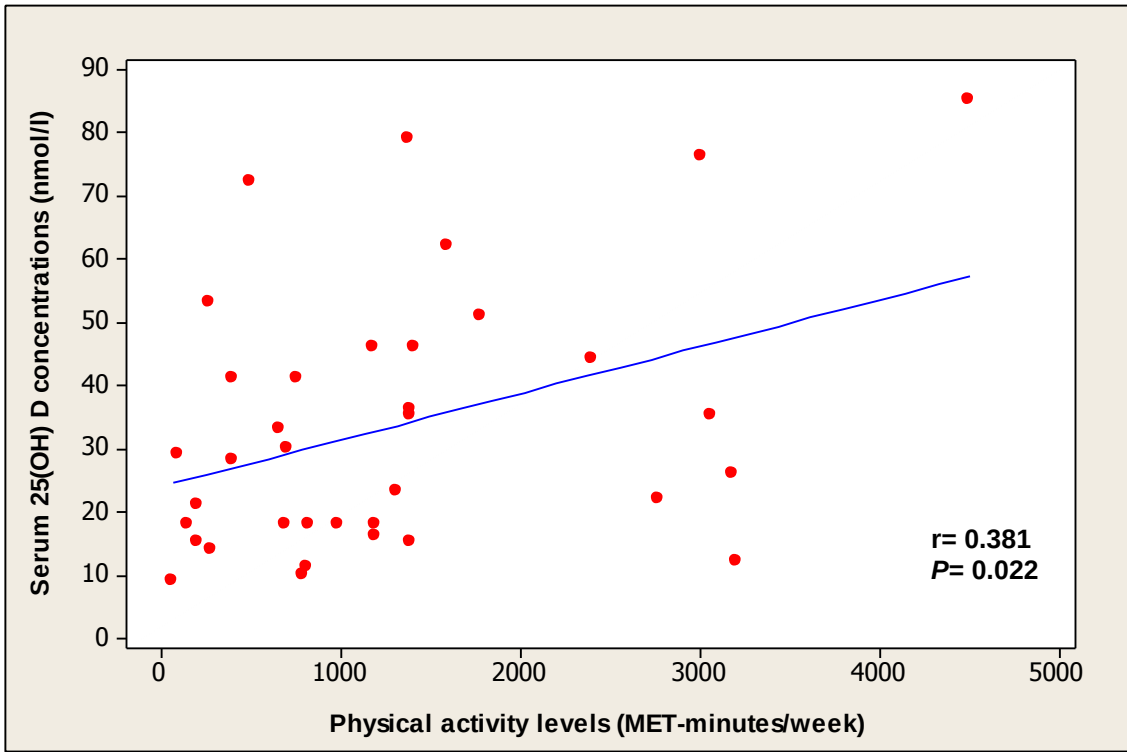

Figure 5. 8 Correlation between serum 25(OH)D concentrations and physical activity levels in total sample (phase1-week12)

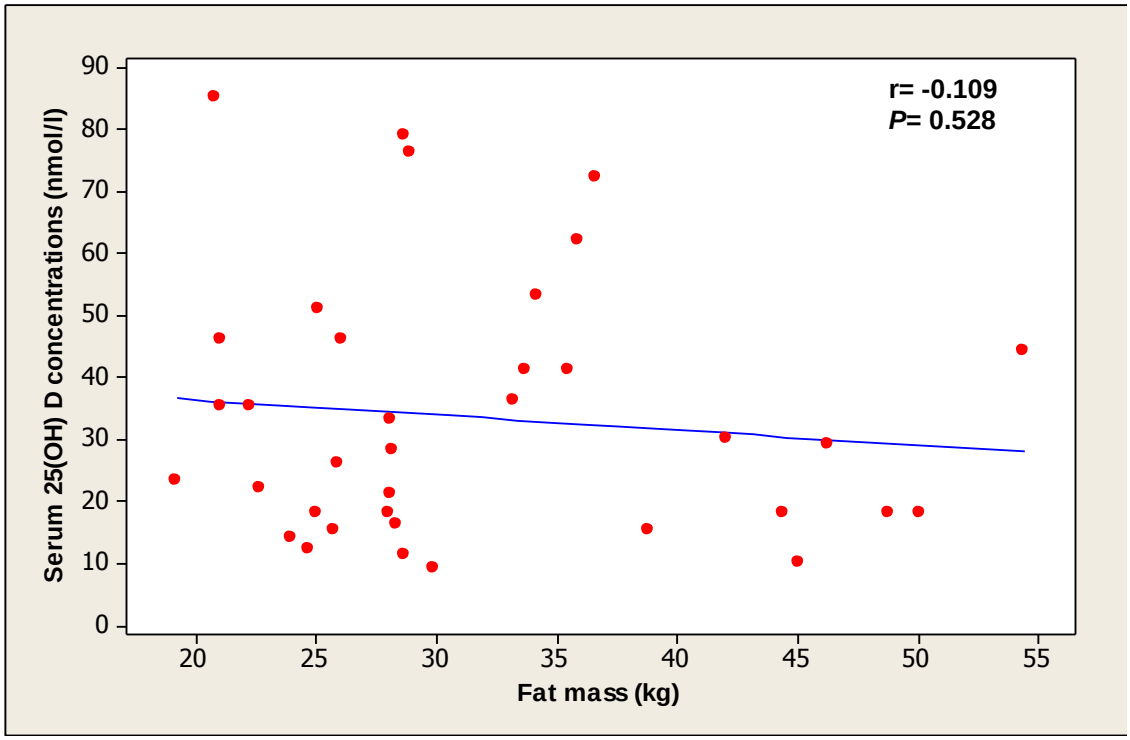

Figure 5. 9 Correlation between serum 25(OH)D concentrations and fat mass in total sample (phase1-week12)

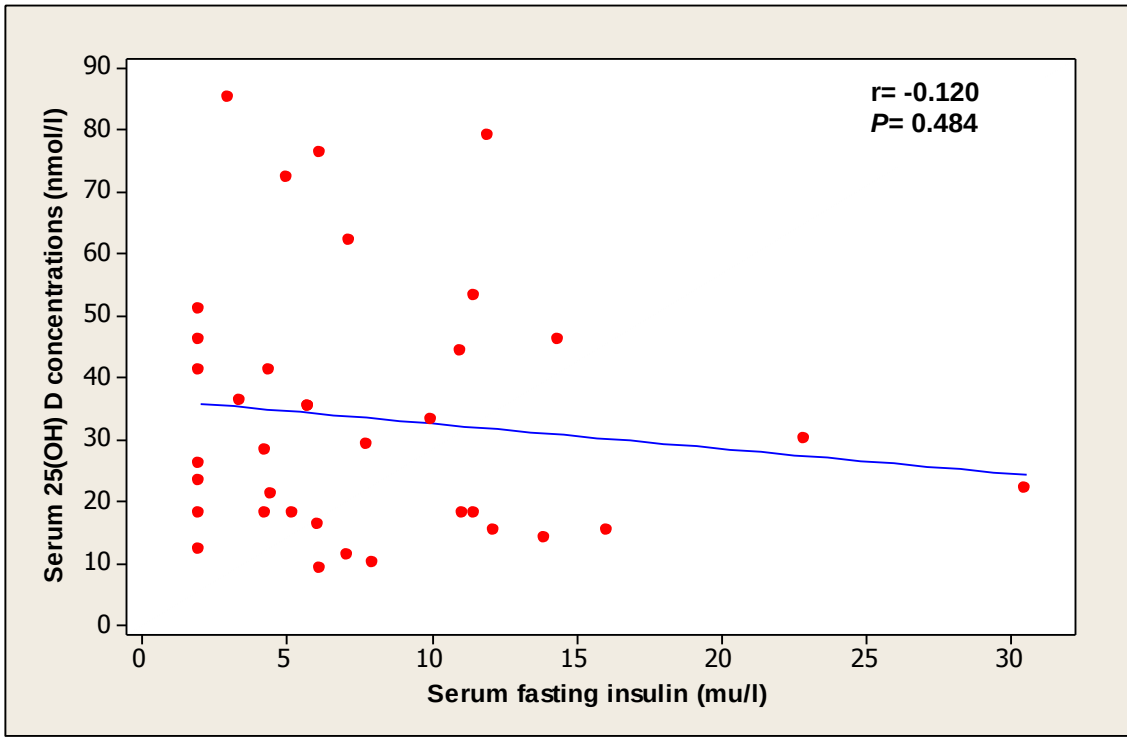

Figure 5. 10 Correlation between serum 25(OH)D concentrations and serum fasting insulin in total sample (phase1-week12)

5.12.5 Characteristics before and after fat mass reduction (week 24) (n=36)

Table 5.10 shows the characteristics of the subjects who lost fat mass and subjects who lost no fat mass. After the body fat reduction programme of 12 weeks, a total of 17 subjects (47.2%) lost ≥1 kg of fat mass, and 19 subjects (52.8%) remained the same or gained fat mass.

Since not all subjects lost fat mass they were split into two groups of roughly equal numbers of those who lost fat mass and those who didn't for comparison. In those that lost fat mass it can be seen in table 5.1, that the mean amount of weight lost was 3.1kg and mean fat loss was 2.6kg, compared to a loss of 0.8kg of weight and a gain of 0.6kg of fat in the group who lost no weight. Although the amount of weight lost was small this would allow the groups to be compared.

5.12.5.1 Characteristics of subjects who lost ≥ 1kg fat mass (week 24) (n =17)

After starting the weight reduction programme, 3 males and 14 females lost ≥ 1 kg fat mass. A decrease of body weight of 3.7%, BMI of 3.8%, waist circumference of 3.6%, hip circumference of 2.6%, body fat percentage 4.5%, and fat mass of 8.2% were achieved. Also fat free mass and total body water slightly decreased. The serum concentrations of 25(OH)D increased by 4.7%; whereas serum fasting insulin stayed at the same level in week 13 and week 24 of the study (Table 5.10).

5.12.5.2 Characteristics of subjects who lost no fat mass (week 24) (n=19)

Among those completing the study, 6 males and 13 females lost no fat mass after 12 weeks of the fat reduction programme. The percentage of body fat and fat mass increased, while fat free mass and total body water decreased However, serum 25(OH)D concentrations and serum fasting insulin were increased by 23.3% and 19.7%, respectively (Table 5.10).

Table 5.10 Sample characteristics before and following fat mass reduction advice (n=36)

	Lost fat mass (n=17)					Lost no fat mass (n=19)				
Age (y)	45.1±11.2					41.2±8.5				
Gender	18% males					32% males				
	January wk 12	**Mar. /Apr. wk 24**	**Change in mean wk 12-24**	**% change**	***P* value**	**January wk 12**	**Mar. /Apr. wk 24**	**Change in mean wk 12- 24**	**% change**	***P* value**
Body weight (kg)	82.9±12.2	79.8±11.6	-3.1	-3.7	0.452	86.5±16.2	85.7±16.2	-0.8	-0.9	0.877
BMI (kg/m^2)	30.4±3.4	29.2±3.2	-1.1	-3.8	0.327	31.6±5.1	31.3±5.3	-0.3	-0.9	0.868
WC (cm)	93.1±10.3	89.7±9.8	-3.4	-3.6	0.338	95.4±12.0	94.1±12.3	-1.3	-1.4	0.741
HC (cm)	109.5±7.0	106.6±6.2	-2.9	-2.6	0.213	111.4±9.4	110.1±9.1	-1.3	-1.1	0.677
WHR (cm)	0.85±0.07	0.84±0.07	-0.01	-0.9	0.759	0.86±0.09	0.85±0.1	-0.01	-0.3	0.935
Body fat (%)	38.9±6.9	37.1±7.0	-1.8	-4.5	0.462	35.6±7.4	36.5±7.7	+0.9	+2.7	0.700
Fat mass (kg)	32.3±8.4	29.7±7.9	-2.6	-8.2	0.350	31.0±10.1	31.6±10.6	+0.6	+1.8	0.866
FFM (kg)	50.5±9.3	50.0±9.3	-0.5	-0.9	0.888	55.4±11.0	54.0±10.7	-1.4	-2.5	0.693
TBW(kg)	37.0±6.8	36.7±6.8	-0.3	-0.9	0.889	40.6±8.0	39.6±7.8	-1.0	-2.5	0.695
25(OH)D (nmol/l)	37.3±21.2	39.0±23.1	+1.7	+4.7	0.818	30.1±20.5	37.1±24.5	+7.0	+23.3	0.346
SFI (mu/l)	7.2±3.8	7.2±4.6	0.0	0.0	0.971	6.4±4.5	7.7±4.0	+1.3	+19.7	0.391
Vitamin D intake (µg/d)	2.4±1.7	-	-	-	-	2.5±2.2	-	-	-	-

Data are expressed as mean ±SD

According to a two sample t-test no significant difference was found in 25(OH)D concentrations between the subjects who lost >1 kg of fat mass and who lost no fat mass in week 12 and week 24 (Figure 5.11). This was perhaps not surprising considering the small amount of fat loss actually achieved by the subjects.

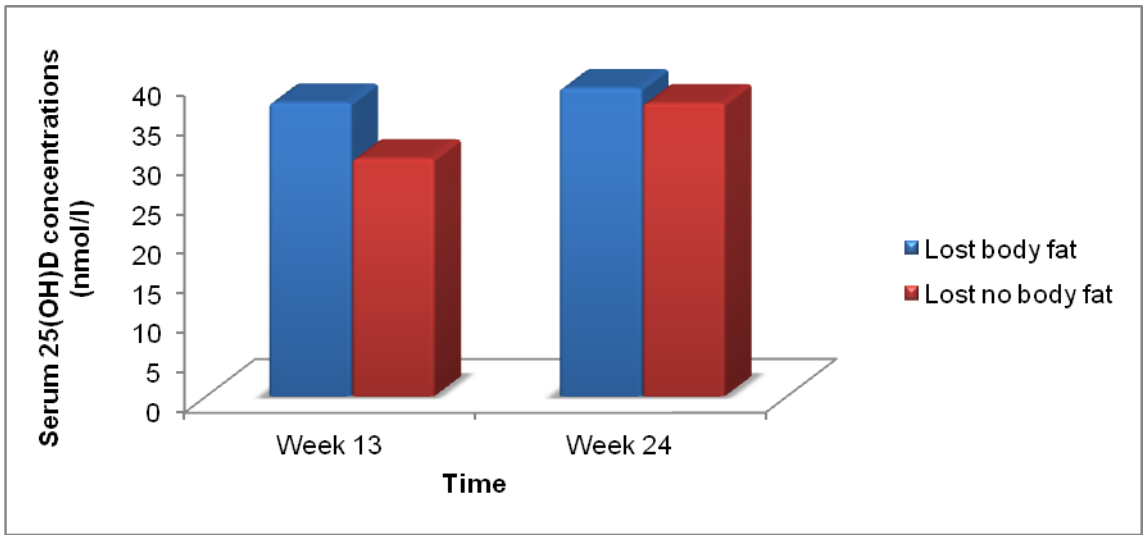

Figure 5. 11 Serum 25(OH)D concentrations in subjects lost and lost no fat mass in phase 2 (week12-24)

5.12.5.3 Physical activity levels following body fat mass reduction advice

Figure 5.12 shows self-reported physical activity levels of the study sample in week 24. No significant differences in physical activity levels were observed between week 1, 12 and 24 of the study sample. Nevertheless, the percentages of highly active and moderate activity subjects were increased from 16.7% and 47.2% at week 12 to 19.4% and 58.3% at week 24, respectively. In contrast, the percentage of low activity subjects was decreased from 36.1% at week 12 to 22.2% at week 24.

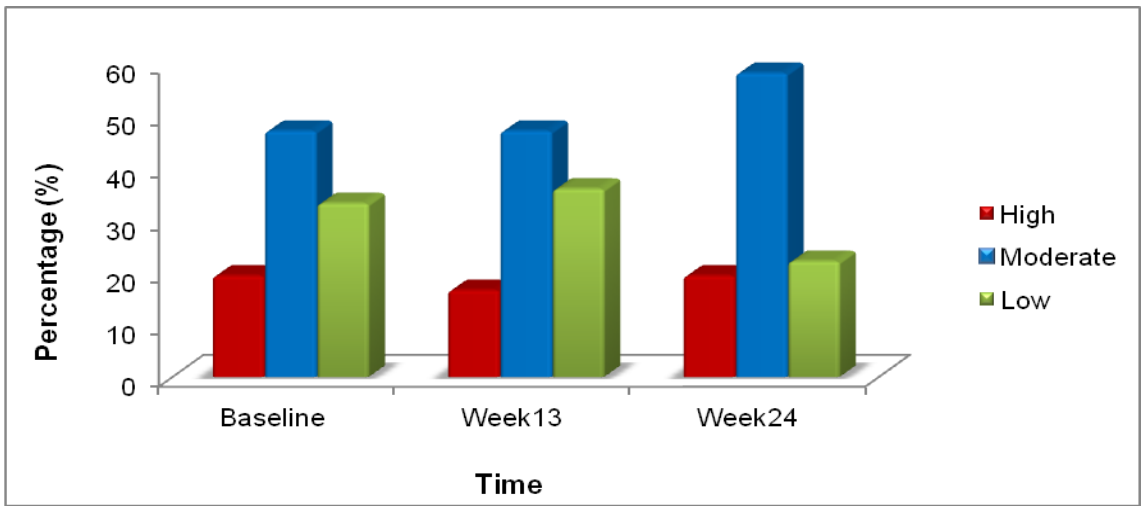

Figure 5. 12 Physical activity levels of the study sample at week1, 12 and 24

Figure 5.13 shows self-reported physical activity levels of the study sample following fat mass reduction advice in phase 2. The chi-square with two degree of freedom (x^2 2) showed no significant differences in physical activity levels between the subjects who lost fat mass and the subjects who lost no fat mass at week 24. Among the “lost body fat“ group 23.5% of the subjects were highly active, 70.6% and 5.9% were of moderate and low activity, respectively. While in “lost no body fat” group 15.8%, 47.4% and 36.8% of the subjects were considered as high, moderate and low activity, respectively.

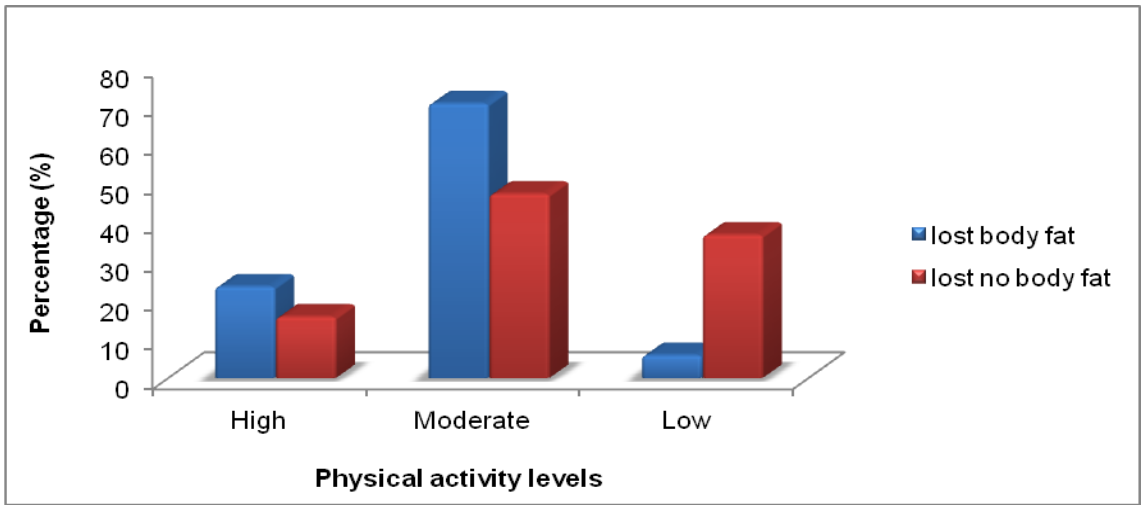

Figure 5. 13 Physical activity levels following body fat reduction advice

5.12.5.4 Correlation between 25(OH)D and other characteristics of the study sample (week 24)

Table 5.11 shows Pearson correlation coefficient analysis results for the whole study sample (n=36) after body fat reduction advice (week 24). There were significant negative correlations between serum 25(OH)D concentrations and waist circumference (r= -0.377, *P*=0.024) (Figure 13) and a marginally significant negative correlation between 25(OH)D and waist-hip ratio (r= -0.325, *P*=0.053) (Figure 5.14).

No significant correlations were found between 25(OH)D and both fat mass (Figure 5.15) and serum fasting insulin (Figure 5.16).

SFI correlated positively with body weight (r= 0.343, *P*=0.041), waist circumference (r= 0.604, *P*=0.0001), and waist-hip ratio (r= 0.658, *P*=0.0001) (Table 5.11).

Table 5.11 Correlation coefficient between 25(OH)D and other characteristics of the study sample at week 24 (n=36)

	SFI (mu/l)	**BW** (kg)	**BMI** (kg/m^2)	**WC** (cm)	**HC** (cm)	**WHR** (cm)	**BF** (%)	**FM** (kg)	**FFM** (kg)	**Physical activity**
25(OH)D (nmol/l)	-0.096 *P*= 0.579	-0.188 *P*= 0.273	0.283 *P*= 0.095	**-0.377** ***P*= 0.024**	-0.166 *P*= 0.335	-0.325 *P*= 0.053	-0.023 *P*= 0.894	-0.127 *P*= 0.459	-0.149 *P*= 0.387	0.215 *P*= 0.207
SFI (mu/l)		**0.343** ***P*= 0.041**	0.035 *P*= 0.838	**0.604** ***P*= 0.0001**	0.081 *P*= 0.639	**0.658** ***P*= 0.0001**	0.111 *P*= 0.518	0.272 *P*= 0.109	0.235 *P*= 0.167	-0.204 *P*= 0.234
BW (kg)			0.301 *P*= 0.074	**0.859** ***P*= 0.0001**	**0.765** ***P*= 0.0001**	**0.460** ***P*= 0.005**	0.186 *P*= 0.277	**0.713** ***P*= 0.0001**	**0.760** ***P*= 0.0001**	-0.137 *P*= 0.426
BMI (kg/m^2)				0.264 *P*= 0.12	0.322 *P*= 0.055	0.081 *P*= 0.638	0.319 *P*= 0.058	**0.426** ***P*= 0.01**	0.032 *P*= 0.851	0.028 *P*= 0.87
WC (cm)					**0.511** ***P*= 0.001**	**0.803** ***P*= 0.0001**	0.111 *P*= 0.518	**0.578** ***P*= 0.0001**	**0.684** ***P*= 0.0001**	-0.156 *P*= 0.365
HC (cm)						-0.091 *P*= 0.598	**0.577** ***P*= 0.0001**	**0.855** ***P*= 0.0001**	0.294 *P*= 0.081	-0.113 *P*= 0.512
WHR (cm)							-0.275 *P*= 0.105	0.071 *P*= 0.68	**0.588** ***P*= 0.0001**	-0.091 *P*= 0.597
BF (%)								**0.817** ***P*= 0.0001**	**-0.494** ***P*= 0.002**	-0.077 *P*= 0.657
FM (kg)									0.086 *P*= 0.617	-0.326 *P*= 0.052
FFM (kg)										-0.232 *P*= 0.174

SFI= Serum fasting insulin
BW=Body weight
BMI=Body mass index
WC=Waist circumference
HC=Hip circumference
WHR=Waist-hip ratio
BF=Body fat
FM= Fat mass
FFM=Fat free mass

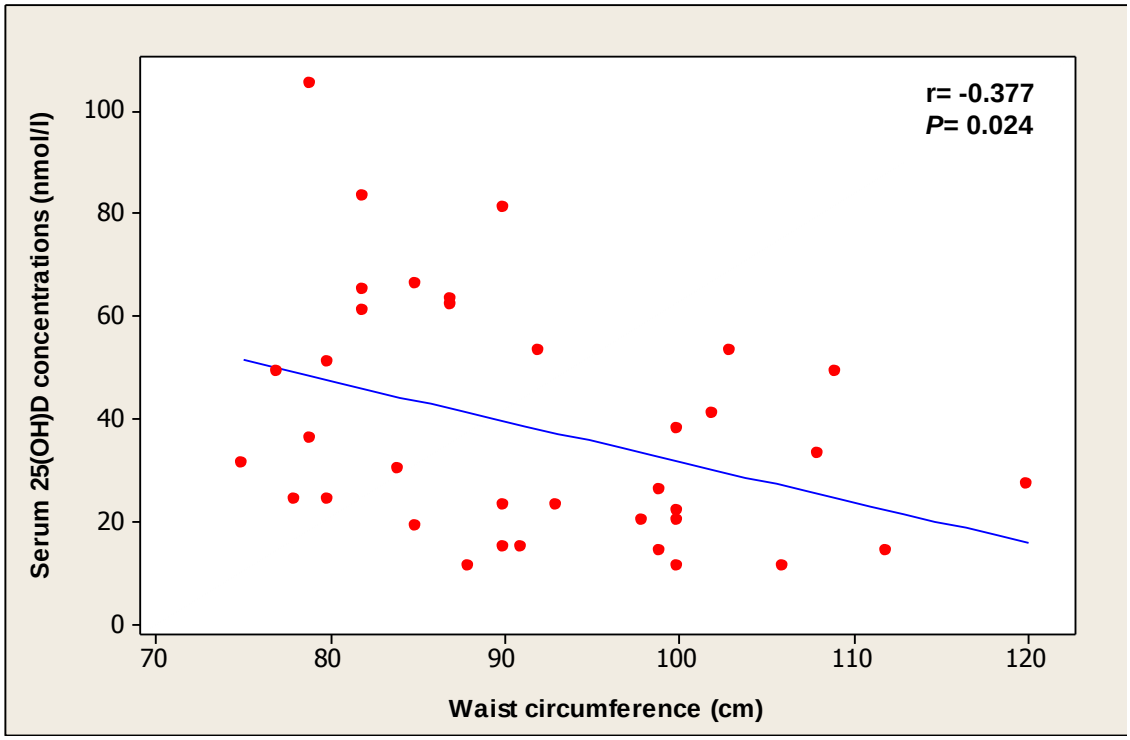

Figure 5. 14 Correlation between serum 25(OH)D concentrations and waist circumference in total sample (phase2-week 24)

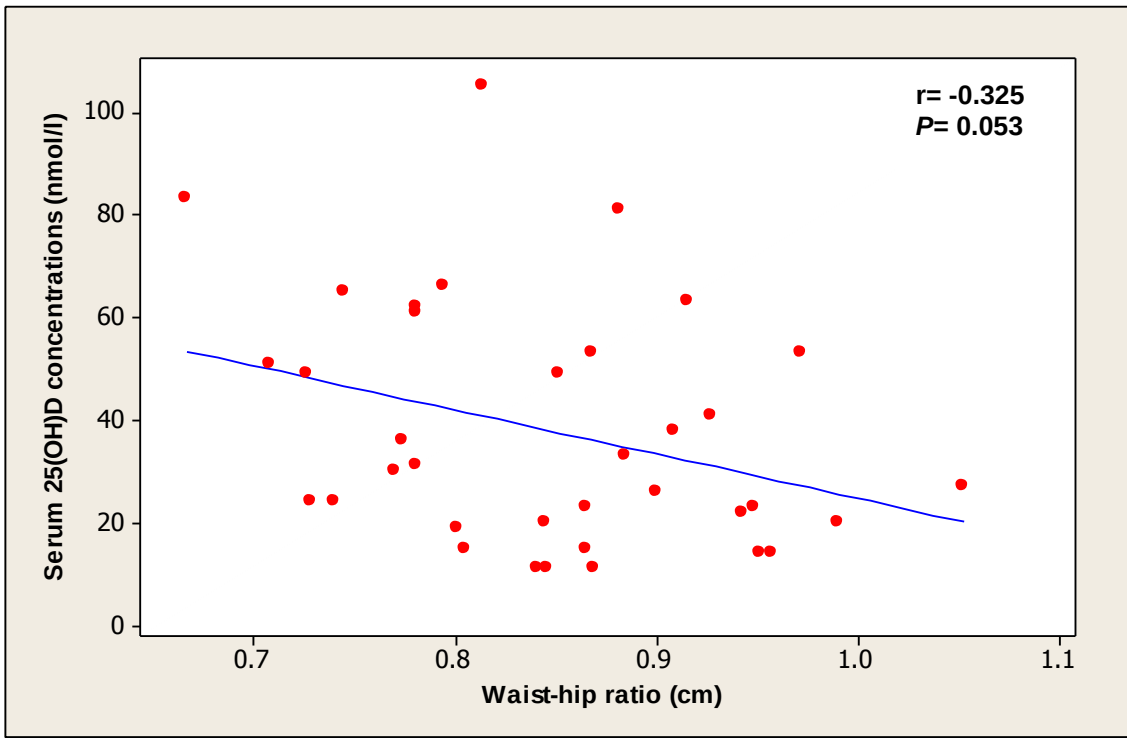

Figure 5. 15 Correlation between serum 25(OH)D concentrations and waist-hip ratio in total sample (phase2-week 24)

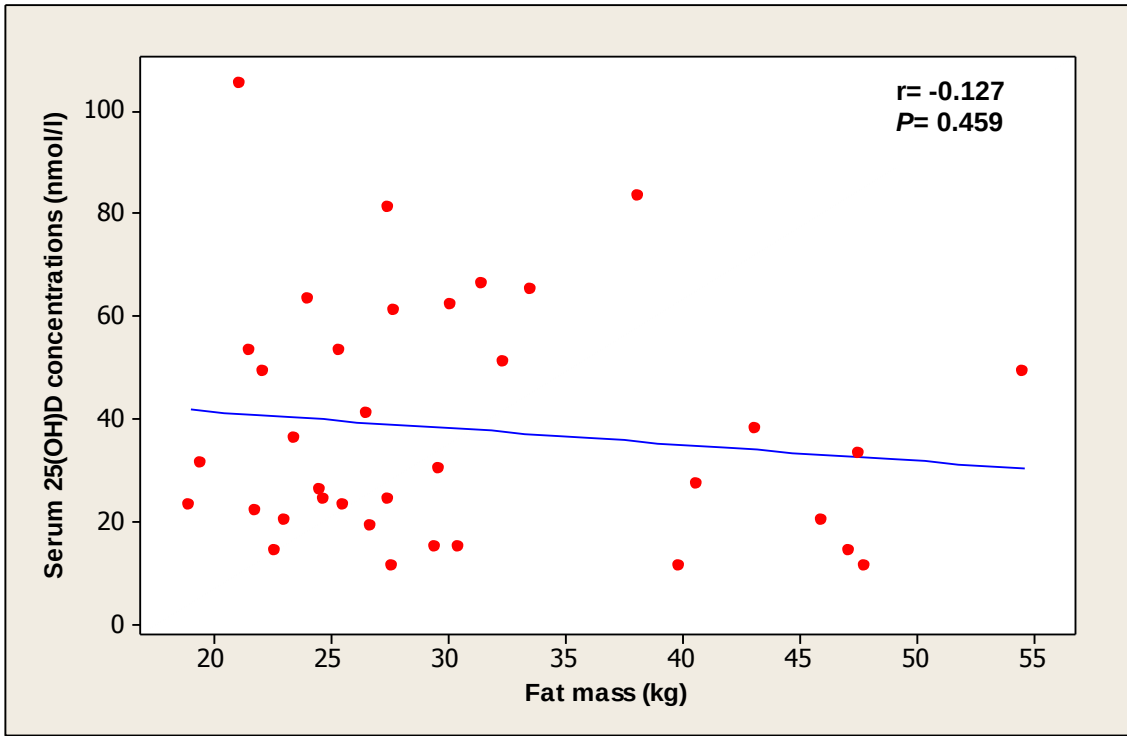

Figure 5. 16 Correlation between serum 25(OH)D concentrations and fat mass in total sample (phase2-week 24)

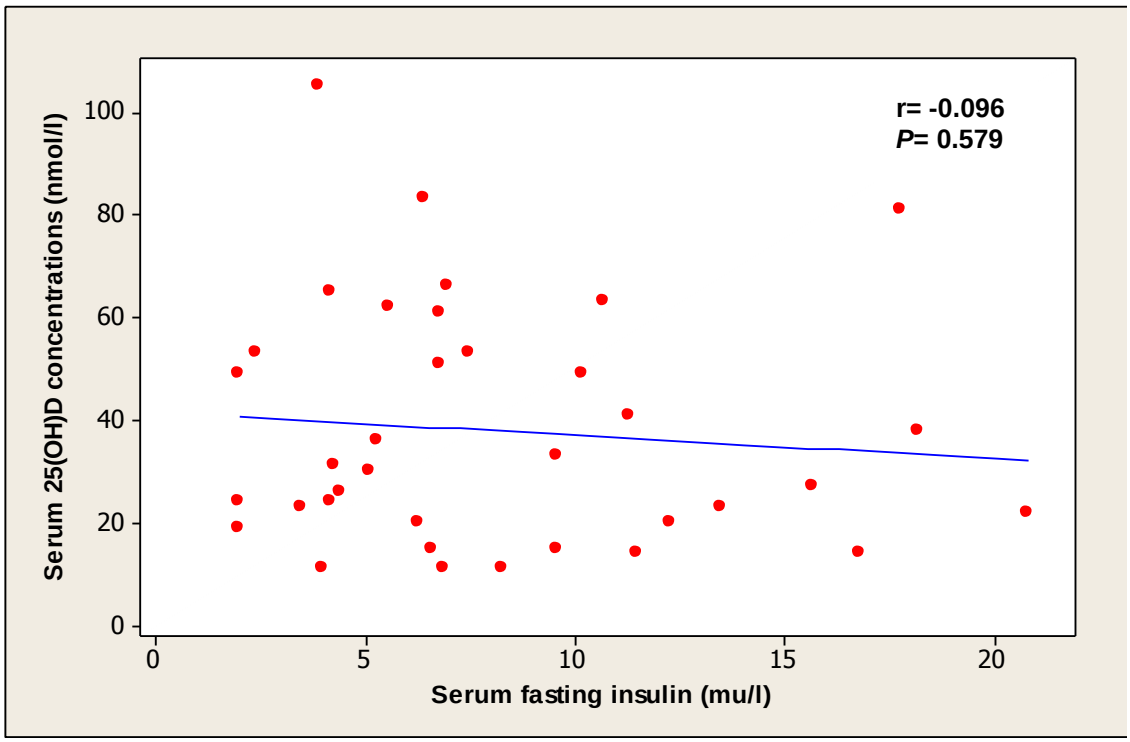

Figure 5. 17 Correlation between serum 25(OH)D concentrations and serum fasting insulin in total sample (phase2-week 24)

5.12.5.5 Correlation between 25(OH)D and other characteristics of subjects who lost fat mass (week24)

Table 5.12 shows Pearson correlation coefficient analysis results for the subjects that lost fat mass (n=17) at week 24. There was a marginally significant negative correlation between serum 25(OH)D concentrations and BMI (r= -0.480, *P*=0.051) (Figure 5.18).

Serum fasting insulin was correlated positively with both waist circumferences and waist-hip ratio (r= 0.488, *P*=0.047 and r= 0.580, *P*=0.015, respectively). On the other hand, serum fasting insulin correlated negatively with physical activity levels (r= -0.526, *P*=0.03) (Table 5.12).

To investigate if the degree of fat loss was related to increase in serum 25(OH)D concentrations, the change in 25(OH)D was correlated with kilograms of fat loss (Figure 5.19). No relationship was found (r = -0.06, *P*=0.798).

Table 5.12 Correlation coefficient between 25(OH)D and other characteristics of subjects lost fat mass at week 24 (n=17)

	SFI (mu/l)	**BW** (kg)	**BMI** (kg/m^2)	**WC** (cm)	**HC** (cm)	**WHR** (cm)	**BF** (%)	**FM** (kg)	**FFM** (kg)	**Physical activity**
25(OH)D (nmol/l)	0.236 *P*= 0.362	-0.245 *P*= 0.343	-0.480 *P*= 0.051	-0.291 *P*= 0.257	-0.422 *P*= 0.091	-0.100 *P*= 0.704	-0.195 *P*= 0.454	-0.321 *P*= 0.21	-0.033 *P*= 0.9	0.040 *P*= 0.879
SFI (mu/l)		0.185 *P*= 0.478	0.191 *P*= 0.463	**0.488** ***P*= 0.047**	-0.008 *P*= 0.976	**0.580** ***P*= 0.015**	0.204 *P*= 0.433	0.265 *P*= 0.303	0.005 *P*= 0.985	**-0.526** ***P*= 0.03**
BW (kg)			**0.708** ***P*= 0.001**	**0.871** ***P*= 0.0001**	**0.698** ***P*= 0.002**	**0.561** ***P*= 0.019**	0.077 *P*= 0.769	**0.604** ***P*= 0.01**	**0.734** ***P*= 0.001**	-0.330 *P*= 0.196
BMI (kg/m^2)				**0.673** ***P*= 0.003**	**0.796** ***P*= 0.0001**	0.292 *P*= 0.255	**0.613** ***P*= 0.009**	**0.892** ***P*= 0.0001**	0.125 *P*= 0.632	-0.418 *P*= 0.095
WC (cm)					**0.510** ***P*= 0.036**	**0.837** ***P*= 0.0001**	0.115 *P*= 0.659	**0.569** ***P*= 0.017**	**0.603** ***P*= 0.01**	**-0.519** ***P*= 0.033**
HC (cm)						-0.019 *P*= 0.944	**0.590** ***P*= 0.013**	**0.841** ***P*= 0.0001**	0.156 *P*= 0.549	-0.130 *P*= 0.619
WHR (cm)							-0.240 *P*= 0.353	0.122 *P*= 0.642	**0.597** ***P*= 0.011**	-0.478 *P*= 0.052
BF (%)								**0.837** ***P*= 0.0001**	**-0.617** ***P*= 0.008**	-0.304 *P*= 0.235
FM (kg)									-0.097 *P*= 0.711	-0.445 *P*= 0.073
FFM (kg)										-0.032 *P*= 0.902

SFI= Serum fasting insulin
BW=Body weight
BMI=Body mass index
WC=Waist circumference
HC=Hip circumference
WHR=waist-hip ratio
BF=Body fat
FM= Fat mass
FFM=Fat free mass

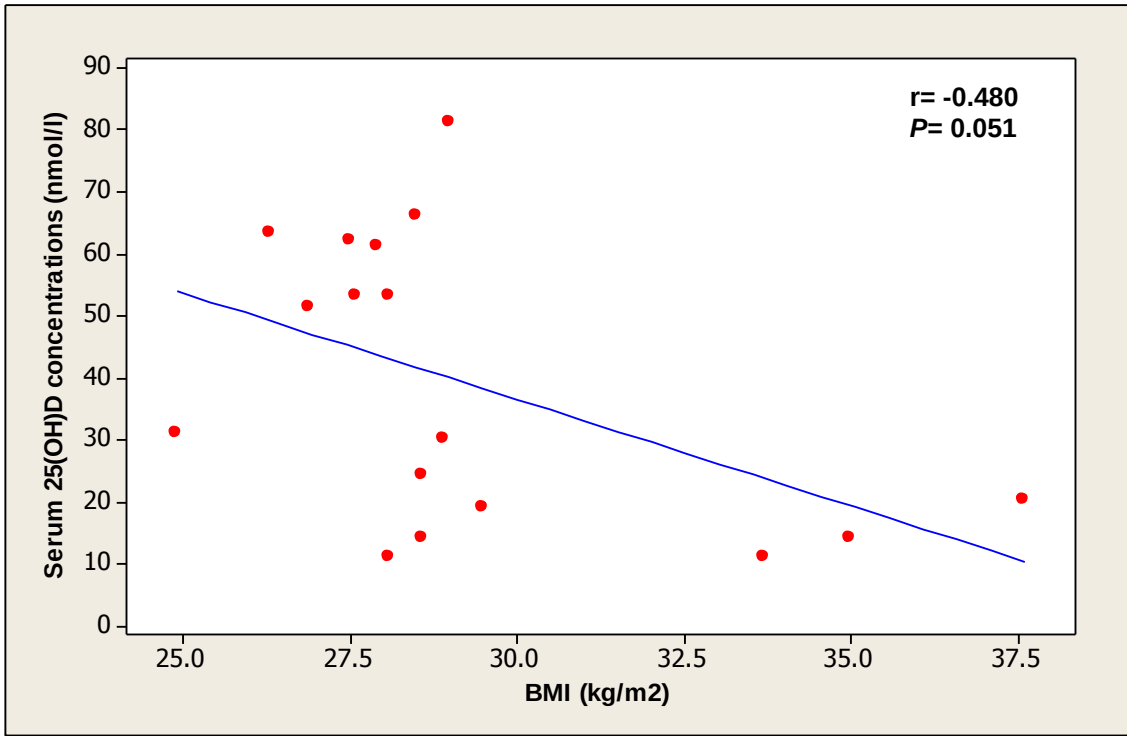

Figure 5. 18 Correlation between serum 25(OH)D concentrations and BMI in subjects lost body fat (phase2-week 24) (n=17)

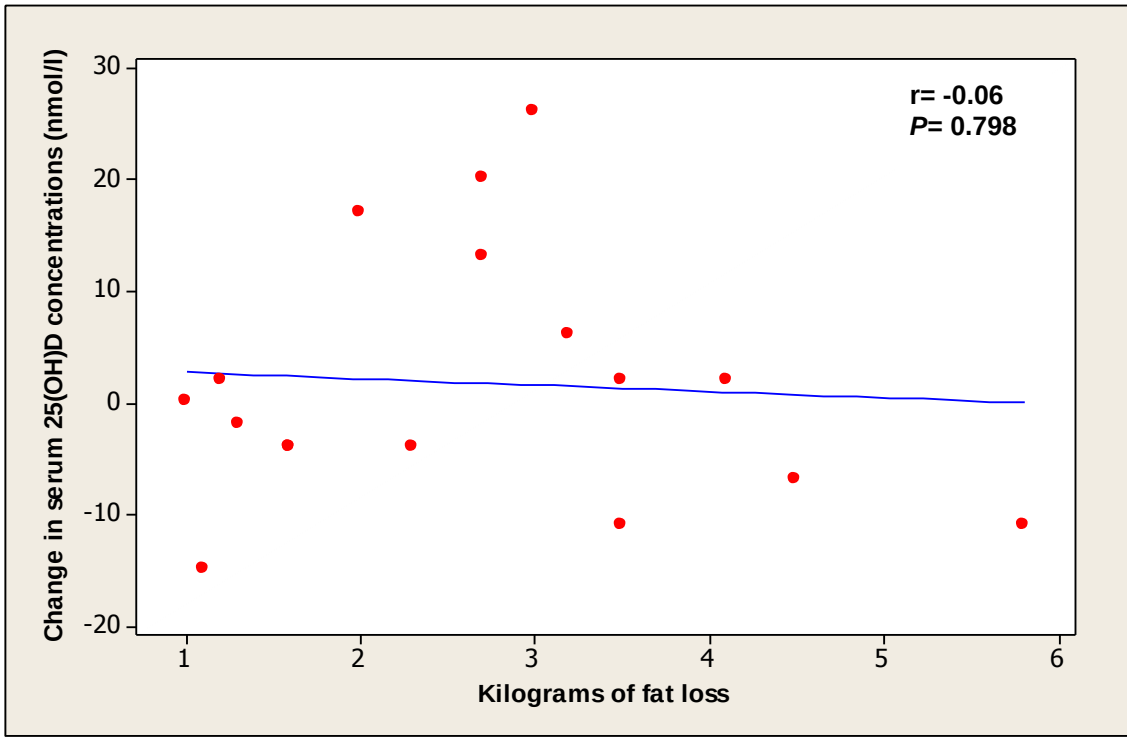

Figure 5. 19 Correlation between change serum 25(OH)D concentrations with kilograms of fat loss (phase2-week24)

5.13 Discussion

5.13.1 Phase 1-observational

Serum (OH) D concentrations and seasonal variation effect

Significant seasonal changes in serum 25(OH)D concentrations were documented in both the Middle Eastern (35.7%) (*P*=0.02) and Caucasian groups (29.1 %) (*P*=0.01) with levels higher at the end of summer than in winter. This reflects the influence of sunlight exposure in summer as solar UVB radiation becomes significantly vitamin D effective (Webb et al. 2010). While, in winter; negligible vitamin D synthesis is possible in the UK after October (Webb et al. 1988, Webb and Engelsen 2006). Several studies in British populations have reported a marked seasonal variation, with the mean serum 25(OH)D concentrations being higher for blood samples collected in summer compared with winter (Hyppönen and Power 2007, Hirani et al. 2009, Webb et al. 2010). For this reason the period for assessing the effect of weight loss on serum 25(OH)D concentrations was undertaken when minimal changes could be attributed to sun exposure during January to March and there was no correlation between activity and serum 25(OH)D concentrations during this time. This is likely to be due to the limited sunlight during winter in the UK.

The Middle Eastern participants had lower serum 25(OH)D concentrations (25.5±11.5 nmol/l) than the Caucasian subjects (59.2±24.5 nmol/l) in both summer (*P*=0.0001) and winter (16.4±5.4 nmol/l and 42.0±20.5 nmol/l, respectively) (*P*=0.0001). Vitamin D sufficiency in summer and vitamin D insufficiency in winter were found among Caucasian sample, while severe vitamin D deficiency was indicated in summer and winter among the Middle Eastern sample. It is important to reveal that no differences in vitamin D status could be detected between Middle Eastern females who wear a headscarf and those who don't as both cover up arms and legs and show only hands and

faces. Moreover, it was just 3 Middle Eastern females (50%) that were wearing a headscarf in this study and 3 who were not and so it was too small a group to compare statistically. It has been indicated that ethnic minority populations (non-white) have lower serum 25 (OH)D concentrations than white British population (Pal et al. 2003, Hirani et al. 2009). These studies have not reported the ethnicity of their non-white subjects, so it is uncertain if any were Middle Eastern. In the current study the most likely reason for the low 25 (OH)D status is the Middle Eastern subject's dress style as most reported covering skin from sun whereas dietary intake was no different to Caucasians. Less than quarter of Middle Eastern subjects reported using sunscreen compared to three quarters of the Caucasians and so it is unlikely that this could account for the low serum 25(OH)D. Numerous studies documented low serum 25(OH)D concentrations among Middle Eastern immigrants in different countries other than the UK (For further discussion, see chapter 2).

5.13.2 Phase 2-intervention

5.13.2.1 Correlations between 25(OH)D and other variables (week 12-24)

- **Body composition measurements**

Serum 25(OH)D concentrations have been shown to be correlated negatively to indices of abdominal obesity such as waist circumference (Snijder et al. 2005, McGill et al. 2008, Cheng et al. 2010, Tzotzas et al. 2010, Forsythe et al. 2012). Moreover, another study indicated that serum 25(OH)D concentrations were highest in the lowest waist circumferences group in non-diabetic subjects aged 40–69 years (Forouhi et al. 2008); this finding was confirmed by the results of the present study for the whole sample, which indicated that serum 25(OH)D concentrations were inversely correlated with waist circumference (r= -0.409,

P=0.013) and waist-hip ratio (r= -0.417, *P*=0.011) at the start of phase 2 (week12). Furthermore, the concentrations of serum 25(OH)D remained significantly inversely correlated with waist circumference (r= -0.377, *P*=0.024) and marginally inversely correlated with waist-hip ratio (r= -0.325, *P*=0.053) at week 24. These findings probably indicate that it was the visceral distribution of body fat and not simply the total body fat that induced a decrease in the serum concentrations of vitamin D. Further evidence for the importance of visceral fat is that no significant correlations were seen between 25(OH)D concentrations and hip circumference in week 12 or week 24, or between serum 25(OH)D and total fat mass or percentage of body fat at either time point.

A study conducted by McGill *et al* (2008), found similar results: there was an inverse association of serum 25(OH)D concentrations with waist circumference, but not with percentage of body fat (r = -0.14, *P*=0.03) in 250 adults in Auckland, New Zealand. The sample recruited into the study had a BMI28-50 kg/m^2, were not currently using weight loss agents nor participating in commercial weight loss programmes, and had a desire to lose weight and were therefore comparable to this study. Thus it would seem to strengthen the idea that serum 25(OH)D only relates to abdominal fat and not total fat.

- **Physical activity**

Our findings of a positive correlation between serum 25(OH)D concentrations and physical activity levels (r= 0.365, *P*=0.029) at week 12 was consistent with other studies which documented that physical activity is associated with increased vitamin D status (Scragg et al. 1992, Scragg et al. 1995, Brock et al. 2007, Looker 2007, van Dam et al. 2007, Freedman et al. 2008, Scragg and Camargo 2008, Foo et al. 2009, Brock et al. 2010a, Brock et al. 2010b,

Kluczynski et al. 2011). The mechanism by which physical activity increases serum 25(OH)D concentrations remains speculative. Physical activity could also just be a surrogate measure for healthier lifestyle, and less body fat (IARC 2008) since significant negative correlations were found between physical actively levels and BMI (r= -0.411, *P*=0.013), hip circumferences (r= -0.351, *P*=0.036), percentage of body fat (r= -0.427, *P*=0.009) and body fat mass (r= -0.425, *P*=0.01) at week 12. These findings are in agreement with the results of the Food Standards Agency (2007), which stated that, as BMI increased in adults physical activity levels decreased. On the other hand, no significant correlation was seen between waist circumference and physical activity levels in week 12 (r= -0.242, *P*=0.155) or in week 24 (r= -0.156, *P*=0.365). Our result is consistent with another study who stated that changes in sports/exercise and daily routine activity appeared to have less influence on waist circumference. Moreover, physical activity may contribute more to preventing increases in the overall amount of fat than preventing redistribution of that fat (Sternfeld et al. 2004). Furthermore, a significant negative correlation was seen between physical activity levels and waist circumference (r= -0.519, *P*=0.033) in subjects who lost fat mass after weight reduction advice. This finding is consistent with the Food Standard Agency (2007), which documented that, decreased physical activity levels are associated significantly with a raised waist circumference.

At week 24 of the study, no significant correlations were found between serum 25(OH)D concentrations and physical activity levels in the total sample (r= 0.215, *P*=0.207) nor in subjects who lost fat mass after the body fat reduction advice (r= 0.040, *P*=0.879). Both correlations, between physical activity and serum 25(OH)D concentrations, of week 12 and 24 are positive, but only at

week 12 were significant. This trend of association is with what is expected between the two variables.

- **Dietary intake of vitamin D**

Dietary intake of vitamin D has consistently been reported as a determinant of serum 25(OH)D concentrations (Brock et al. 2007, van Dam et al. 2007, Foo et al. 2009, Hirani et al. 2009, Brock et al. 2010a, Brock et al. 2010b). This finding was confirmed by the results of the present study, which found that serum 25(OH)D concentrations were positively correlated with dietary intake of vitamin D (r= 0.459, *P*=0.005) at week 12.

- **Fasting serum insulin**

Serum 25(OH)D concentrations have been reported to be negatively correlated with insulin levels (Tzotzas et al. 2010). Nevertheless, the concentrations of serum 25(OH)D in our study did not correlate significantly with serum fasting insulin in week 12 (r= -0.120, *P*=0.484), in week 24, (r= -0.096, *P*=0.579) nor in subjects who lost body fat (r= 0.236, *P*=0.362). This finding was also found in a study of 51 African-American adolescents (r= -0.074, *P*=0.61) (Ashraf et al. 2009).This could be due to the short period of the intervention study of 12 weeks of body fat reduction programme.

It was also found that serum fasting insulin was associated with both waist circumference (r= 0.441, *P*=0.007) and waist-hip ratio (r= 0.530, *P*=0.001) in week 12, and in week 24 and in subjects who lost fat mass. This is consistent with a study conducted by Cigolini and others (1991), which indicated significant correlations between serum insulin and waist circumference and serum insulin and waist-hip ratio in healthy women. This might be, due to the well-known

observation that, central obesity confers a greater risk of insulin resistance and type 2 diabetes (McTernan et al. 2002).

In addition, a marginally significant negative correlation was observed between SFI and physical activity levels (r= -0.328, *P*=0.051).This finding is consistent with Kriska et al. (2001) and Chlebowski et al. (2004) . This is likely due to the fact that physical inactivity is a major risk factor for the development of type 2 diabetes. Two of the key mechanisms underlying this relationship between physical activity and diabetes involve the influence of physical activity on improving insulin sensitivity (Lindgärde et al. 1983).

5.13.2.2 Effect of fat loss on serum 25(OH)D concentrations

The effect of fat mass loss (kg) has been examined on serum 25(OH)D concentrations and serum insulin in Middle Eastern and Caucasian individuals. After 12 weeks of fat mass reduction advice; diverse total body fat loss outcomes were seen; a decrease in total body fat mass of 8.2% was achieved in 17 (47.2%) subjects who lost ≥1 kg fat mass. On the other hand, a total of 19 (52.8%) subjects remained the same or gained fat mass.

At the start of phase 2 (week 12), serum 25(OH)D concentrations and serum fasting insulin levels were higher in “lost fat mass group” than in “lost no fat mass group” but these differences were not significant. Daily intakes of vitamin D (µg/d) were similar between the two groups. The fat mass reduction advice of 12 weeks did not result in significant fat mass loss and therefore this did not significantly influence those metabolic parameters in the “lost fat mass group” and there was no significant difference in the serum 25(OH)D at week 24 between those that lost fat mass and those that did not. However, an increase in 25(OH)D was observed of 4.7% in “lost fat mass group” and 7.0% in “lost no fat mass group”. This was most likely due to increased sun exposure which

increased the opportunities for synthesis of vitamin D through skin (Table 5.13) since there was no fat loss and the diet was unlikely to have changed substantially since week 12. It can be seen in Table 5.13 that during March and April 2011 solar radiation had increased substantially from the winter months and it is this that is likely to have increased the serum vitamin D in some of the participants. Ideally sun exposure should have been monitored in the participants or the final serum samples taken before solar radiation increased as spring approached. However sufficient time was needed for weight loss to occur.

Table 5.13 Day length and solar radiation in Plymouth over the study period

Date	**Day length***	**Solar radiation****
October 2010	10hr 55min	763 J/cm^2
January 2011	8hr 41min	233 J/cm^2
March-April 2011	12hr 56min	1467 J/cm^2

*Sunrise and sunset in Plymouth (2012)
**SODA (2012)

No correlation was seen between fat mass loss (kg) and increase in serum 25(OH)D concentrations. This is likely to be attributed to the small sample and too little fat loss.

5.14 Limitations

There are some limitations to the current study these include:

1- Sampling the Middle Eastern population in the South West was difficult due to culture, invasive nature of the study (venepuncture) and total number of subjects available at the University to recruit. This was reflected in the small size of the sample that contributed to this study. In the future it would be of interest to recruit from different centres so that more subjects could be studied.

2- Calculation of sample size required was based on a mean weight loss of 10.4 kg (Tzotzas, et al., 2010) which was a much greater weight loss than achieved by our sample A much larger sample size would probably be required to detect differences in vitamin D levels with the small weight loss achieved by our subjects. Nevertheless, we did manage to detect correlations: vitamin D concentrations were associated with waist circumference and waist: hip as a clinical sign of central obesity.

3- This sample only included Iraqi subjects and therefore does not represent other Middle Eastern groups. However, this is the first study to assess vitamin D status in Iraqi people. The published literature does not allow a comparison between different ethnic groups and therefore it is not possible to comment on whether significant differences would be found between different groups of Middle Eastern people.

4- Ideally it would have been of benefit to measure dietary intake at the end of the study (week 24) to indicate whether participants were following dietary advice but this would have been difficult as many of the participants were reluctant to complete the food diary for the third time and found the number of appointments for venepuncture, anthropometrics and questionnaires

difficult to attend. Rather than risk obtaining poor quality data it was reluctantly decided to omit this later measurement of diet.

5- The intervention period of 12 weeks of body fat reduction programme was relatively short and a longer period as demonstrated by Tzotzas and others (2010) would have been preferable. However, the longer the intervention period the greater the risk of changes in serum concentrations of vitamin D due to seasonal changes (greater exposure to sunlight). A much larger study with greater number of controls would have been required.

6- Underreporting of habitual food intake had an effect on the results for energy and macronutrient intake and both would be higher especially since subjects were overweight and obese. Nevertheless, underestimation may only have affected certain meal types such as high fat snacks (Poppitt et al. 1998). Snack foods are unlikely to have contributed significantly to dietary vitamin D. The estimation of dietary intake using a food diary is known to underestimate vitamin D (Sowers and Wallace 1986). However, this was largely attributed to failure to record supplement use. In this study individuals using supplements were not included.

7- The use of Dietplan6 software programme may have introduced some minor errors as it does not contain all the food items and brand names of Middle Eastern traditional diets. However, care was taken to analyse recipes using basic ingredients to provide as accurate nutritional analysis as possible.

8- Exposure to sunlight was not measured in this study and this is known to influence serum 25(OH)D concentrations. The study was carried out over the course of the winter months when the level of exposure to sunlight is limited.

5.15 Conclusion

In conclusion, vitamin D deficiency is very common in Middle Eastern adults living in the UK and shows seasonal variation; vitamin D concentrations were associated with waist circumference and waist-hip ratio as a clinical sign of central obesity; small amounts of fat loss following diet and exercise advice did not improve serum 25(OH)D levels.

CHAPTER 6: Dietary sources of vitamin D and dietary assessment methods

6.1 The most important dietary sources of vitamin D

6.1.1 The 24 hour dietary recall method

Daily intake of vitamin D for Middle Eastern people (n=28) was assessed using three 24 hour dietary recalls (Chapter 4, study 2). The estimation of vitamin D intake was performed using the nutritional analysis software CompEat. The most important dietary source of vitamin D from this data was eggs which were reported more frequently than other vitamin D food sources: they were the main source of vitamin D for 15 participants. Meat was the second most important source of vitamin D and lamb was the most frequently eaten meat source (Table 6.1). Dairy products were the third most important dietary source of vitamin D. Cakes, biscuits, pastries, pancakes and mayonnaise were a source of vitamin D as they contain eggs and/ or margarine. However, they were only a main source for two participants and a second source for three more participants. For example the two participants with cakes, biscuits and pastries as a main source of vitamin D obtained 0.5 and 0.3 µg/d from these foods.

Table 6.1 The most important dietary sources of vitamin D reported in 24 hour dietary recall

Food items	Number of participants									
	Source 1	Source 2	Source 3	Source 4	Source 5	Source 6	Source 7	Source 8	Source 9	Source 10
Eggs	15	7	4	2						
Lamb	5	8	7	1						
Lamb liver				1	1					
Beef	2	4	4	3	4	1	1	1		
Chicken	1	2	2	4	4	6	1	3	1	
Milk	1	1		1	3		1			
Yogurt		1	2	2	3		1	1		
Cheese		1	1	7	2	5	7	1	1	2
Cream			2			1	1		1	
Margarine	1		1							
Cakes, Biscuits and Pastries *	2	2	2	3	2					
Pancake and Mayonnaise **		1	1	1		2		1	1	

* Foods contain eggs and /or margarine
** Foods contain eggs

6.1.2 The prospective Food Diary method

- **Middle Eastern participants**

The daily intake of vitamin D for the Middle Eastern people (n=12) was assessed using a 3 day food diary in October 2010 (week 1) and in January 2011 (week 12) (Chapter 5, study 3). The estimation of vitamin D intake was performed using the nutritional analysis software Dietplan6.

Tables 6.2 and 6.3 summarize the most important dietary sources of vitamin D reported by Middle Eastern participants by using Dietplan6. The data showed that, similar to the 24 hour recall data, eggs were the most important dietary sources of vitamin D as they were the main source of vitamin D for 6 participants. As previous, meat was the second most important source of vitamin D; dairy products were the third dietary sources of vitamin D followed by cakes, biscuits and pastries. Unlike the 24 hour recall data, 1 participant reported intake of canned tuna as a first source of vitamin D and another participant reported intake of breakfast cereals as source of vitamin D.

- **Caucasian participants**

Daily intake of vitamin D for the Caucasian people (n=24) was assessed using food diary in October 2010 (week 1) and in January 2011 (week 12) (Chapter 5, study 3). The estimation of vitamin D intake was performed using the nutritional analysis software Dietplan6. Tables 6.4 and 6.5 summarize the most important dietary sources of vitamin D reported by Caucasian participants by using Dietplan6. The data indicated that the main dietary sources of vitamin D among Caucasians were eggs, oily fish, canned tuna, margarine, breakfast cereals and dairy products. Moreover, cakes, biscuits, pastries, mayonnaise and salad cream were sources for small amounts of vitamin D as they contain margarine and eggs.

Table 6.2 The most important dietary sources of vitamin D reported in food diary (Middle Eastern- week1)

Food items	Number of participants									
	Source 1	Source 2	Source 3	Source 4	Source 5	Source 6	Source 7	Source 8	Source 9	Source 10
Eggs	6	6								
Canned tuna	1									
Lamb	2	2	2	3	2	1			1	
Beef	1	1	1		1	2				
Chicken			6	4	1	3	2			1
Milk			1		1			1		
Yogurt				1	1	1	2			
Cheese			1	2	3	3	2	4	1	1
Margarine							2			
Breakfast cereals				1						
Cakes, Biscuits and Pastries *	2	2	1	1	2		1			

* Foods contain eggs and/or margarine

Table 6.3 The most important dietary sources of vitamin D reported in food diary (Middle Eastern-week12)

Food items	Number of participants								
	Source 1	Source 2	Source 3	Source 4	Source 5	Source 6	Source 7	Source 8	Source 9
Eggs	6	4	2						
Canned tuna	1								
Lamb	1	4	3	1	1			1	
Beef	1	1							
Chicken	1	2	1	1	2	1	1		
Milk					1	1			
Yogurt	1	1			1	2			
Cheese		1	1	2	1	1	2	1	1
Cream				2	2		1		
Breakfast cereals	1								
Cakes, Biscuits and Pastries *			2	1	1		1		

* Foods contain eggs and/or margarine

Table 6.4 The most important dietary sources of vitamin D reported in food diary (Caucasian-week1)

Food items	Number of participants									
	Source 1	Source 2	Source 3	Source 4	Source 5	Source 6	Source 7	Source 8	Source 9	Source 10
Eggs	5	1	2	3	2	1				
Canned tuna	4	2	1							
Oily fish	5				1					
Lamb		1	2	1						
Beef	2	7	3	1	3	2	1			
Chicken	1	1	5	4	4	1	2	1		
Turkey		1								
Pork*	1	3	4	1	1	2	3		1	
Liver			1							
Milk		1	1		1	2			1	
Yogurt					1	2		1		
Cheese		1	1	10	3	3	4	3	1	1
Margarine	3		1							
Butter			1	3	1	4	3	3		
Cream						1		2	2	
Breakfast cereals		1	1							
Cakes, Biscuits and Pastries **	4	5	4	1	2		1	1		
Mayonnaise and Salad cream ***				1	6	1	3	2	1	1

* Including bacon rashers and sausages
**Foods contain eggs and/or margarine
*** Foods contain eggs

Table 6.5 The most important dietary sources of vitamin D reported in food diary (Caucasian-week12)

Food items	Number of participants								
	Source 1	Source 2	Source 3	Source 4	Source 5	Source 6	Source 7	Source 8	Source 9
Eggs	5	2	1	2		1			
Canned tuna	5								
Oily fish	4	3		1	1				
Lamb			1						
Beef	1	4	5	2	1				
Chicken		3	3	2	6	4	1	1	
Turkey	1					1			
Pork*	1	4		2	3	1			
Yogurt			1	1		1	1	1	
Cheese		1	5	3	7	2			1
Margarine	1	2	2	1					
Butter		1	3	1		4	1		
Cream					1				1
Breakfast cereals	4	1	1						
Cakes, Biscuits and Pastries **	1	2	2	2			1		
Mayonnaise and Salad cream ***		1		4	1	3	3		

* Including bacon rashers and sausages
**Foods contain eggs and/or margarine
*** Foods contain eggs

6.2 Comparison of dietary vitamin D assessment methods

6.2.1 Background

Errors in food reporting and quantification can vary with the type of dietary methodology. This thesis examined three methods of dietary assessment: Food Frequency Questionnaire (FFQ), repeat 24 hour recalls and a 3 day food diary, and so it is useful to compare the three methods to see if they estimate a similar intake of vitamin D within the same participants.

6.2.2 Aim

The purpose of this chapter was to evaluate the FFQ with 31 items for rapid assessment of vitamin D intake in Middle Eastern adults living in the UK (used in study 1).

6.2.3 Objective

Compare the assessment of vitamin D intake by using the FFQ to assessment by 24 hour dietary recall and food diary.

6.2.4 Subject recruitment and methods

Details of subject's recruitment and methods to estimate vitamin D intake by using FFQ, 24 hour dietary recall and food diary have been described in chapter 3, chapter 4 and chapter 5, respectively.

6.2.5 Sample characteristics

6.2.5.1 FFQ and 24 hour dietary recall

A total of 22 subjects (11 males and 11 females) completed the FFQ and 24 hour dietary recall. The mean age was 37.2±11.7 y (range: 22-70 y). All the subjects were of Iraqi decent, and lived in Plymouth and London.

6.2.5.2 FFQ and food diary

A total of 9 subjects (4 males and 5 females) completed the FFQ and food diary. The mean age was 34.8±4.3 y (range: 29-42 y). All the subjects were of Iraqi decent, and lived in Plymouth.

6.2.5.3 The 24-hour dietary recall and food diary

A total of 5 subjects (2 males and 3 females) completed a 24-hour dietary recall and food diary. The mean age 32.4±3.5 y (range: 29-36 y). All the subjects were of Iraqi decent, and lived in Plymouth.

6.2.6 Comparison of vitamin D intake

The Pearson correlation coefficients between the FFQ and 24 hour dietary recall, between the FFQ and the food diary, as well as between 24 hour dietary recalls and food diary, are shown in tables 6.6, 6.7, and 6.8.

When we compared intakes from the FFQ and the mean of three 24 hour dietary recall, there was a significant positive correlation (r=0.469, *P*=0.028). However, the t-test showed that the mean intakes from FFQ (2.5±1.2 µg/d) were significantly higher than those from the 24 hour dietary recall (1.5±0.8 µg/d) (*P*=0.035) (Table 6.6).

Similarly a significant positive correlation were found between the FFQ and food diary (r=0.672, *P*=0.048). But this time the t-test showed no significant differences (*P*=0.068) of mean daily intake of vitamin D from FFQ (2.1±1.2µg/d) and food diary (1.3±0.5 µg/d) (Table 6.7).

For the small number who undertook both the 24 hour recalls and the food diary, no significant correlation was found (r=0.428, *P*=0.472) (Table 6.3). Moreover, no significant differences (*P*=0.834) were found between the mean intakes from the 24 hour dietary recall (2.0±0.7 µg/d) and food diary (2.1±1.1 µg/d) (Table 6.8).

Table 6.6 Comparison of vitamin D intakes estimated from FFQ and 24 hour dietary recall (n=22)

Vitamin D intake (µg/d)		Pearson correlation coefficients	*P* value correlation	*P* value t-test
FFQ	24 hour recalls			
2.5±1.2	1.5±0.8	0.469	0.028	0.035

Intakes are expressed as mean ±SD

Table 6.7 **Comparison of vitamin D intakes estimated from FFQ and food diary (n=9)**

Vitamin D intake (µg/d)		Pearson correlation coefficients	*P* value correlation	*P* value t-test
FFQ	Food diary			
2.1±1.2	1.3±0.5	0.672	0.048	0.068

Intakes are expressed as mean ±SD

Table 6.8 **Comparison of vitamin D intakes estimated from 24 hour dietary recall and food diary (n=5)**

Vitamin D intake (µg/d)		Pearson correlation coefficients	*P* value correlation	*P* value t-test
24 hour recalls	Food diary			
2.0±0.7	2.1±1.1	0.428	0.472	0.834

Intakes are expressed as mean ±SD

6.2.7 Discussion

6.2.7.1 The most important dietary sources of vitamin D

The detailed information collected from 24 hour dietary recalls (analysed by CompEat) in chapter 4 and food diary (analysed by Dietplan6) in chapter 5 on foods consumed shows that Middle Eastern subjects were consuming similar vitamin D sources in the two studies. Reported intakes of eggs were high compared to other vitamin D sources. It has been found that eggs were the main source of vitamin D for more than half of the subjects took part in 24 hour dietary recalls study. In addition, eggs were the main source of vitamin D for half of Middle Eastern subjects who participated in week 1 and week 12 of weight loss study (chapter 5). Results obtained from chapter 3 showed that eggs were widely consumed among Middle Easten participants. Moreover, another study reported that eggs are an important food item in the Middle East diet (Al-Khateeb and Al-Gelban 2008).

Meat was consumed more frequently than fish, and lamb was preferred over beef. Low intake of fish was reported by other studies in the Middle East (Nasreddine et al. 2006, Al-Khateeb and Al-Gelban 2008) and fish is not an important vitamin D source unlike in the British diet.

Although all main food sources of vitamin D reported by Middle Eastern people in chapter 4 and 5 were listed in the FFQ (See appendix 1), some foods which incorporated eggs and/or margarine into recipes such as: cakes, biscuits, pastries, pancakes and mayonnaise were omitted from the food questionnaire. Even though the data showed that vitamin D content in these foods is relatively low, it was observed that the intakes of vitamin D from these foods were a main source in two participants (although only 0.5 and 0.3µg/d). Therefore, the

omission of these foods in the FFQ would not affect the estimation of dietary intake of vitamin D greatly in chapter 3.

On the other hand, it was found that the main dietary sources of vitamin D among Caucasians were eggs, oily fish, canned tuna, margarine and breakfast cereals. The national diet and nutrition survey for adults aged 19 to 64 years (NDNS 2003) also reported that the main sources of vitamin D in the diets of respondents in the UK were fish and fish dishes, meat and meat products, cereals and cereal products and fat spreads.

6.2.7.2 Comparison of dietary assessment methods

- **FFQ and 24 hour dietary recall**

Our study revealed a significant positive correlation (r=0.469, *P*=0.028) between the FFQ and 24 hour dietary recall data of assessment daily intake of vitamin D indicating a positive relationship between the two methods.

The mean intake of vitamin D estimated by FFQ was greater than that estimated by the 24hour dietary recalls; this could be due to the fact that we had overestimated the portion sizes to get an over-estimation of vitamin D intake rather than risk under-estimation. Moreover, FFQ tend to overestimate the consumption of food items (Jonneland et al. 1991, Sichieri and Everhart 1998, Wu et al. 2009) which, in this study, appeared to be true for vitamin D foods and as a consequence may have overestimated vitamin D intake.

- **FFQ and food diary**

In the present study, there were a significant positive correlation (r=0.672, *P*=0.048) between FFQ and food diary data of assessment daily intake of vitamin D; once again this result indicated a positive relationship. However this time there was no significant difference between the two estimations despite the FFQ giving a seemingly higher result. It should be noted that the samples size for this comparison was smaller than previous.

- **The 24 hour dietary recall and food diary**

No significant correlations were found between the 24 hour dietary recall and food diary in estimation daily intake of vitamin. This is likely to be attributed to the small sample (n=5).

6.2.8 Conclusion

Eggs were the greatest food source of vitamin D for the Middle Eastern participant living in the UK and fish is less frequently eaten. Eggs and oily fish were the greatest sources of vitamin D among Caucasians.

We assessed the validity of the food questionnaire for estimating vitamin D intake in Middle East adults by comparison to the use of three 24 hour recalls and a 3 day diet diary. The comparison of the assessment methods revealed a tendency for higher estimation of mean vitamin D intake (µg/d) using the FFQ than the 24 hour dietary recall despite omission of some of the vitamin D containing foods such as cakes and pastries. However, over-estimations of nutrient intakes using the FFQ are a known limitation to this method. The FFQ results were correlated to the other two methods showing that there was a positive relationship. It should be remembered that the FFQ gives an indication of a longer term eating pattern of subjects and the other methods only recorded intakes for 3 days so provides only short term measures.

CHAPTER 7: General discussion

7.1 General findings

Firstly a questionnaire based survey was undertaken with 242 Middle Eastern respondents. A total of 85% of the sample was estimated to have a vitamin D intake < 5 µg /d. Other risk factors for vitamin D insufficiency included covering skin from sunlight (62% males and 84% females); low use of vitamin D supplements (18.5%) and being overweight or obese (49% males and 44% females).

The survey was followed by dietary assessment of 28 Middle Eastern volunteers using repeat 24 hour recall methodology to collect detailed information on dietary habits and nutritional intake. The results of estimated daily intake of vitamin D using 24 hour recall (1.4±0.8 µg/d) concurred with the survey data (3.2±4.4 µg/d) and showed that mean intake of vitamin D was lower than the FAO/WHO (2002) reference value of 5µg/d. In this study, 57.1% of the subjects were overweight and 21.4% were obese.

Finally, due to the high prevalence of overweight and obesity found in the previous work and the fact that obesity is known to adversely affect vitamin D status, overweight participants were recruited to observe the effect of fat loss on vitamin D status. Serum 25(OH)D concentrations were measured in Middle Eastern adults (n=12) and compared to a Caucasian group (n=24). Firstly participants were advised to keep their weight stable so that seasonal changes could be observed between October and January. Between January and April

participants were then advised on weight reduction to observe the effect of fat loss on serum 25(OH)D concentrations.

Vitamin D deficiency (<25 nmol/l) was observed in 67% of the Middle Eastern group in October and 4% of the Caucasian group. This increased to 92% of the Middle Eastern and 21% of the Caucasians in January.

Of the 36 participants, only 17 lost (≥1kg) of fat mass between January and April. No difference was found in serum 25(OH)D concentrations between those that lost fat mass and those that did not and no correlation was found between the amount of fat lost and change in serum 25(OH)D concentrations. In the total sample, no correlation was found between total fat mass and serum 25(OH)D but there was a negative association between serum 25(OH)D concentrations and waist circumference (r= -0.377, *P*=0.024) and waist-hip ratio (r= -0.325, *P*=0.053).

On the other hand, this study shows that our Food Frequency Questionnaire can be used as a valid tool to estimate vitamin D intake among Middle Eastern adults in the UK.

7.2 Suggestions for future research

The large numbers of vitamin D studies generated convincing results that, obesity is inversely related to concentrations of serum 25(OH)D (See Appendix 7), and it is possibly a result of decreased vitamin D bioavailability due to sequestration in fat tissue (Wortsman et al. 2000). Additionally, several researchers found that obesity and BMI have been negatively correlated with the change in serum 25(OH)D concentrations following supplementation (See chapter 2, section 2.5.1) and suggested that adiposity and or/body size may need to be taken into account when determining dietary vitamin D intake

required for optimal status (Barger-Lux et al. 1998, Blum et al. 2008a, Lee et al. 2009b, Forsythe et al. 2012). On the other hand, some studies have found no such association (Canto-Costa et al. 2006, Nelson et al. 2009). Therefore further interventional studies are required to investigate the effect of body composition on serum 25(OH)D concentrations wintertime response to vitamin D supplement (cholecalceferol) in Middle Eastern obese and lean subjects with serum 25(OH)D <25 nmol/l.

The current research investigated factors affecting vitamin D status in Middle Eastern people in the UK. However, the trial that exist in chapter 5 assessed serum 25(OH)D concentrations in overweight and obese Middle Eastern subjects that had a high percentage of body fat (> 20% for males and > 30% for females) (Grodner et al. 2012). It would therefore be useful to assess serum 25(OH)D concentrations in normal weight Middle Eastern to compare vitamin D status with overweight and obese Middle Eastern adults.

7.3 General conclusion

The main strengths of the study included the fact that this is the first comprehensive study in Middle East population living in the UK to compare factors affecting vitamin D status and estimated dietary intake and to assess the prevalence of vitamin D deficiency in Middle Eastern subjects now living in the UK.

This study shed light on the lifestyle and eating habits of Middle Eastern people living in the UK. It has been indicated that, this group avoid sun exposure and abstain from eating foods rich in vitamin D and taking vitamin D supplements; vitamin D intake was generally higher in more educated participants, in those

aged 40-49 y, and those who were obese. However, 85% of the sample had an estimated intake of vitamin D less than the recommended.

In addition, a high number of participants were overweight or obese, and although the obese subjects reported a higher dietary intake, obese individuals may have a lower bioavailability of vitamin D and be at more risk of deficiency. Therefore, they were prone to vitamin D deficiency in winter and summer. It has been identified that, about two third of overweight and obese Middle Eastern individuals were vitamin D deficient (66.6%) and 33.3% were considered as in state of insufficiency during summer. Moreover, seasonal variations were marked in our study with significant differences between summer and winter in serum 25(OH)D concentrations in both Middle Eastern and Caucasian subjects. Additionally, most of both Middle Eastern and Caucasian subjects did not meet the limits of the FAO/WHO (2002) recommendations for vitamin D intake, which are (5 µg/d) for adults 19-50 y including pregnant and lactating women, (10 µg/d) for adults 51-65 y and (15 µg/d) for elderly adults >65 y.

This study also showed that, the concentrations of serum 25(OH)D were clearly associated inversely with waist circumferences and BMI, and positively with physical activity and dietary intake of vitamin D. On the other hand, serum fasting insulin correlated positively with waist circumferences and negatively with physical activity levels. On the other hand, underreporting of energy and nutrients intake is quite common among overweight and obese individuals.

In conclusion, most Middle Eastern people in the UK at risk of vitamin D deficiency as a result of poor dietary and supplementary intake of vitamin D, inadequate sunshine exposure and obesity.

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List of Abbreviations

Abbreviations	Glossary of terms used
%BF	Percentage of Body Fat
µg	Microgramme
1,25(OH) $_2$D	1, 25-dihydroxycholecalciferol
25(OH)D	Serum vitamin D concentrations
ANOVA	Analysis of Variance
BIA	Bioelectrical Impedance Analysis
BMI	Body Mass Index
BMR	Basal Metabolic Rate
BW	Body Weight
CI	Confidence Interval
cm	Centimetre
EI	Energy Intake
FFM	Fat Free Mass
FFQ	Food Frequency Questionnaire
FM	Fat Mass
hr	Hour
HC	Hip Circumference
IPAQ	International Physical Activity Questionnaire
J/cm^2	Joule per square centimeter
kg	Kilogram
LRNI	lower Reference Nutrient Intake
METs	Metabolic equivalent tasks
mu/l	Milliunits per litre
n	Number

nmol/l	Nanomoles per litre
RNIs	Reference Nutrient Intakes
SD	Standard Deviation
SFI	Serum Fasting Insulin
SPF	Sun Protection Factor
TBW	Total Body Water
UK	United Kingdom
UOP	University of Plymouth
UVB	Ultraviolet B photons
VDR	Vitamin D Receptor
Vitamin D_2	Ergocalciferol
Vitamin D_3	Cholecalciferol
WC	Waist Circumference
WHO	World Health Organization
WHR	Waist-Hip Ratio
wk	Week
y	Years

APPENDIX 1:

Food and Health Questionnaire

Food and health questionnaire

Medical information

Please answer the following questions as accurately as possible

Please tick (✔) the box if it applies to you

1- Do you suffer from any of the following?

☐ Osteoporosis ☐ Parathyroid disorder ☐ Intestinal disorder

☐ Liver disease ☐ Kidney disease ☐ Fat malabsorption

☐ Heart disease ☐ Cancer ☐ Hypertension

☐ Type 1 diabetes ☐ Milk allergy and lactose intolerance

☐ Other, please list below:

…….. …….. …….. ……..

…….. …….. …….. ……..

Could you , please take the time to fill in the below questionnaire

2. Do you have a history of vitamin D deficiency? Yes ( ) No ( )

3. List any prescribed or over the counter medications you take or have taken (in last month):

…….. …….. …….. ……..

4. If your family have a history of diseases, please specify below:

…….. …….. ……..

5. Are you pregnant? Yes ( ) No ( ) N/A ( ) If yes, how many weeks? ……

6. Are you breast-feeding? Yes ( ) No ( ) N/A ( )
If so, how old is your infant? ……

7. Do you smoke cigarettes? Yes ( ) No ( )
If yes, on the average, about how many cigarettes a day do you smoke now?

……

8. Do you do outdoor activities with exposure to sunlight (for your job or as leisure time)? Yes ( ) No ( )
If yes, how long do you spend sitting or walking under sunlight each day? …….

9. Do you use sunscreen? Yes ( ) No ( )

If yes, what is the sun protection factor (SPF) number of the sunscreen you use most often?

2 ( ) 8 ( ) 15 ( ) 20 ( ) 30 ( ) 45 ( ) 60 ( )

10. Does your dress style often include? sunglasses ( ) long sleeved shirt ( )

headscarf ( ) veil ( ) hat ( )

11. Height…… Weight ……

(Please indicate units, leave blank if unknown)

Nutritional information

Please answer the following questions as accurately as possible

1. Do you drink alcohol? Yes ( ) No ( )

If yes, how many units* of alcohol do you consume? …….

* refer to the alcohol unit's chart for reference

2. Do you drink beverages that contain caffeine (i.e. coffee, tea, and cola)?

Yes ( ) No ( )

If yes, how often? Seldom/Never…… Weekly…… Daily……..

3. Do you currently take a nutritional supplement (Vitamins/Minerals)?

Yes ( ) No ( ) If yes, how often do you take it?

	Occasionally	**Weekly**	**Daily**
Vitamin D	☐	☐	☐
Vitamin A	☐	☐	☐
Vitamin E	☐	☐	☐
Vitamin B complex	☐	☐	☐
Multivitamin supplement	☐	☐	☐
Cod liver oil	☐	☐	☐
Calcium	☐	☐	☐
Mineral supplement	☐	☐	☐
Other	☐	☐	☐

4. How often you usually eat the following foods? **(Please do not skip any foods)**

Please tick (✔) the box of the following:

	Rarely (never)	Monthly	Weekly			Daily	
			1	2-3 times	5-6 times	once	more
Fish /shellfish							
Salmon	☐	☐	☐	☐	☐	☐	☐
Mackerel	☐	☐	☐	☐	☐	☐	☐
Sardines	☐	☐	☐	☐	☐	☐	☐
Tuna	☐	☐	☐	☐	☐	☐	☐
Shrimp/prawns	☐	☐	☐	☐	☐	☐	☐
Other	☐	☐	☐	☐	☐	☐	☐
Canned fish							
Tuna	☐	☐	☐	☐	☐	☐	☐
Sardines	☐	☐	☐	☐	☐	☐	☐
Other	☐	☐	☐	☐	☐	☐	☐
Meats							
Beef	☐	☐	☐	☐	☐	☐	☐
Lamb /mutton	☐	☐	☐	☐	☐	☐	☐
Goat	☐	☐	☐	☐	☐	☐	☐
Pork and ham	☐	☐	☐	☐	☐	☐	☐
Chicken liver	☐	☐	☐	☐	☐	☐	☐
Lamb liver	☐	☐	☐	☐	☐	☐	☐
Beef liver	☐	☐	☐	☐	☐	☐	☐
Pork liver	☐	☐	☐	☐	☐	☐	☐
Sausage	☐	☐	☐	☐	☐	☐	☐
Hamburger	☐	☐	☐	☐	☐	☐	☐
Poultry							
Chicken	☐	☐	☐	☐	☐	☐	☐
Turkey	☐	☐	☐	☐	☐	☐	☐
Eggs							
Whole eggs	☐	☐	☐	☐	☐	☐	☐
Yolks only	☐	☐	☐	☐	☐	☐	☐
Whites only	☐	☐	☐	☐	☐	☐	☐
Milk /dairy products							
Whole Milk	☐	☐	☐	☐	☐	☐	☐
Semi skim milk	☐	☐	☐	☐	☐	☐	☐
Skim milk	☐	☐	☐	☐	☐	☐	☐
Cheese	☐	☐	☐	☐	☐	☐	☐
Butter	☐	☐	☐	☐	☐	☐	☐
Margarine	☐	☐	☐	☐	☐	☐	☐
Cream	☐	☐	☐	☐	☐	☐	☐
Yogurt	☐	☐	☐	☐	☐	☐	☐
Breakfast cereals	☐	☐	☐	☐	☐	☐	☐

Personal Information:

Code No. ☐

Answer by the ticking (FAO/WHO/UNU) in the correct circle:

Gender: Male ◯ Female ◯ **Occupation:** ……..…….

Educational Qualifications: Primary ◯ Secondary ◯ Diploma ◯
Bachelor ◯ Postgraduate ◯ other ◯

Date of Birth: (mm/dd/yy) ……..…….. **Nationality:** ……..……

Country of Birth: ……..…… **How long you have been in the UK?** ……

Religion: Muslim ◯ Christian ◯ Mandaean ◯ Yezidi ◯
Druze ◯ Bahai ◯ Jewish ◯ other ◯

Ethnic origin: Arab ◯ Kurd ◯ Assyrian ◯ Turkoman ◯
Armenian ◯ Berber ◯ Persian ◯ other ◯

- **Please, take a moment to fill in any questions that you may have skipped**

- **Thank you very much for your co-operation and time** ☺

- **If you are happy to be contacted about helping us in the next stage of this study, please fill in the information below:**

Name:

Last: …….. First: ……..

Postal Address: ……..……..……
……

……..……..……

Phone number (Including dialling code):

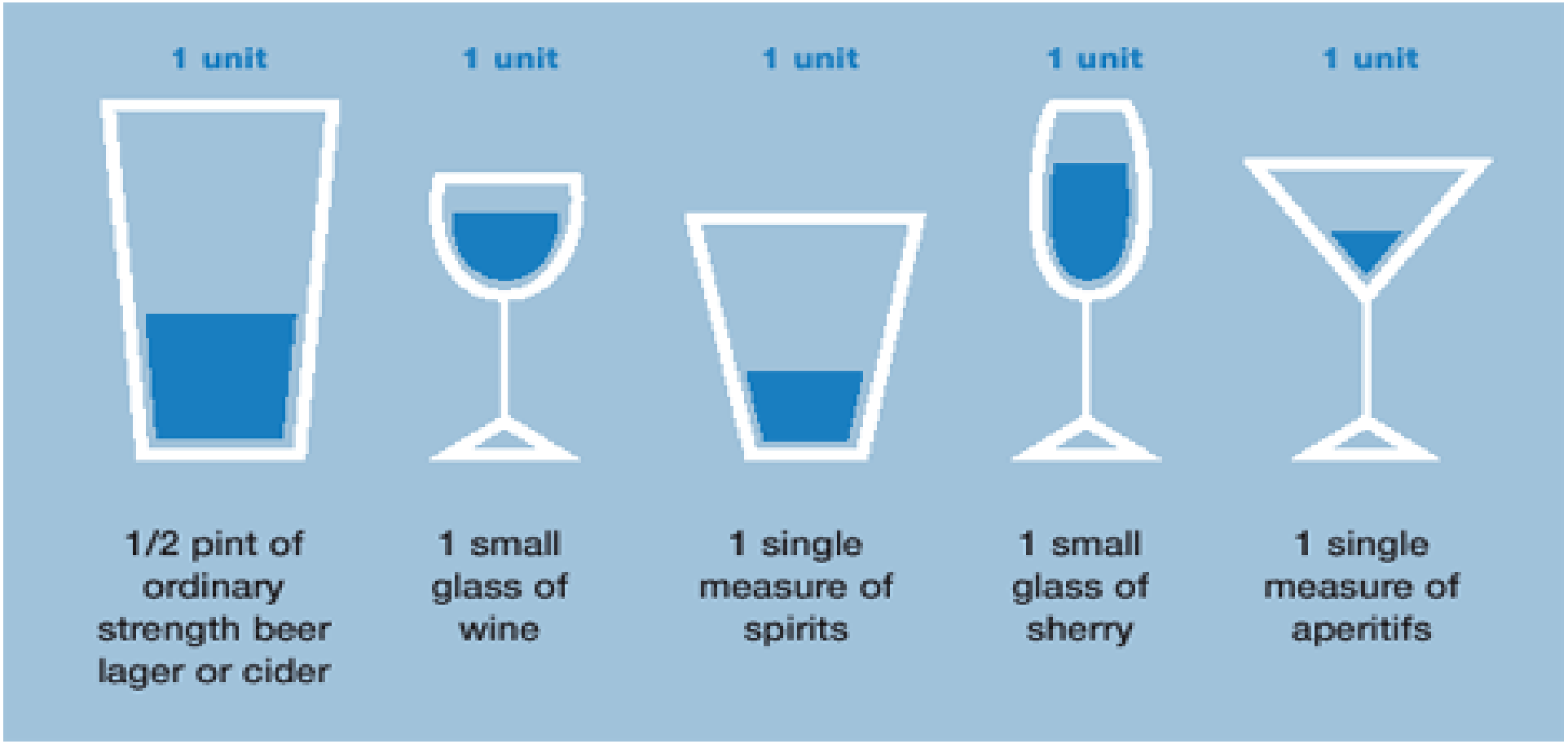
Landline: …….. Mobile: ……..

We may need to contact you:

Please tell us what time is most convenient to phone you……

E-mail: …….. **Code No.** ☐

Alcohol Unit Chart



APPENDIX 2:

Consent Form (Chapter 3)

UNIVERSITY OF PLYMOUTH

FACULTY OF SCIENCE

CONSENT TO PARTICIPATE IN RESEARCH PROJECT

Name of Principal Investigator: Wassan Ahmed

Title of Research: Vitamin D intake and other risk factors for vitamin D insufficiency in Middle Eastern people living in the UK: A comparison of cultural and ethnic groups

Brief statement of purpose of work:

In this project you will be asked to fill in a food and health questionnaire. You will be asked questions about any diseases that you or your immediate family have, any medication that you take and information regarding your diet and lifestyle. This is so that we can investigate the different eating habits, lifestyle and health of people from the Middle East living in the UK. At a later date, some participants may be contacted again and asked to participate in further studies regarding their vitamin D levels which you will be free to accept or refuse at that time.

The objectives of this research have been explained to me:

- I understand that I am free to withdraw from the research at any stage, and ask for my data to be destroyed if I wish.
- I understand that my anonymity is guaranteed, unless I expressly state otherwise.
- I understand that the Principal Investigator of this work will have attempted, as far as possible, to avoid any risks, and that safety and health risks will have been separately assessed by appropriate authorities (e.g. under COSSH regulations).

Under these circumstances, I agree to participate in the research

Name: ……..

Signature: …….. **Date:** ……..

APPENDIX 3:

24 Hour Dietary Recall

24 hour dietary recall

DIET HISTORY
Breakfast: **Mid-morning:** **Lunch:** **Mid-afternoon:** **Evening meal:** **Supper:**

CHECK LIST/COMMENTS

Fish:

Meat/liver:

Poultry:

Eggs:

Milk whole/ semi skimmed/skimmed/ cheese/ butter/ cream/ yoghurt: whole-low fat

Breakfast cereals:

Vitamin & mineral supplement:

LIFESTYLE/RISK FACTORS:

Age:

Weight:

Height:

Name:

Signature:

Date:

APPENDIX 4

Consent Form (Chapter 4)

UNIVERSITY OF PLYMOUTH

FACULTY OF SCIENCE

CONSENT TO PARICIPATE IN RESEARCH PROJECT

Name of Principal Investigator: Wassan Ahmed

Title of Research: Dietary intake of vitamin D and calcium in Middle Eastern people living in the UK

Brief statement of purpose of work:
In this project you will be asked to tell the interviewer about all the food/drink you consumed within last 24 hrs including: what food/drink was consumed, how much was consumed, time it was consumed, how it was prepared, how was it served and the specifics of the food (low fat etc). You will be asked to do this on 3 separate occasions at your convenience.

The objectives of this research have been explained to me:

I understand that I am free to withdraw from the research at any stage, and ask for my data to be destroyed if I wish.

I understand that my anonymity is guaranteed, unless I expressly state otherwise.

I understand that the Principal Investigator of this work will have attempted, as far as possible, to avoid any risks, and that safety and health risks will have been separately assessed by appropriate authorities (e.g. under COSSH regulations).

Under these circumstances, I agree to participate in the research

Name: ……..

Signature: …….. **Date:** ……..

APPENDIX 5

Consent Form (Chapter 5)

UNIVERSITY OF PLYMOUTH

FACULTY OF SCIENCE AND TECHNOLOGY
CONSENT TO PARICIPATE IN RESEARCH PROJECT

Name of Principal Investigator: Wassan Ahmed

Title of Research: The relation between body fatness and vitamin D status of Middle Eastern people and Caucasians living in the South West of the UK

Brief statement of purpose of work:
The aim of this study is to investigate the relationship between vitamin D status and body fatness in Middle Eastern people and Caucasians.
You will be asked to give fasting blood samples to assess vitamin D levels at week 0, 12 and 24 of the study to find out the effect of season (3 months) and the body fat loss (3 months) on vitamin D status. Blood samples will be taken in a quiet clinical room set aside for this purpose by a trained phlebotomist.
You will be asked to keep a food diary for 3 days and physical activity will be estimated via a questionnaire. Also weight, height, waist circumference, hip circumference and body fat will be measured. These measurements will be taken at week 0, 12, 16 and 20 and 24 of the study. We will also collect some information on sun exposure and medications in a questionnaire.
After 3 months you will be given advice for a healthy diet for weight loss and advice to increase your physical activity. This will happen at week 12, 16 and 20 with a qualified dietitian.
At end of the study you will be counselled on any changes that could still be made to make your diet healthier. Vitamin D levels will be revealed at end of the study and appropriate advice on diet/supplements given.

The objectives of this research have been explained to me:

I understand that I am free to withdraw from the research at any stage, and ask for my data to be destroyed if I wish.
I understand that my anonymity is guaranteed, unless I expressly state otherwise.
I understand that the Principal Investigator of this work will have attempted, as far as possible, to avoid any risks, and that safety and health risks will have been separately assessed by appropriate authorities (e.g. under COSSH regulations)

Under these circumstances, I agree to participate in the research

Name: ……

Signature: …… **Date:** ……

APPENDIX 6

Participant Information Leaflet

Researcher:
Wassan Ahmed
School of Biomedical
and Biological Sciences
University of Plymouth
Wassan.ahmed@plymouth.ac.uk
Tel: 01752 584695

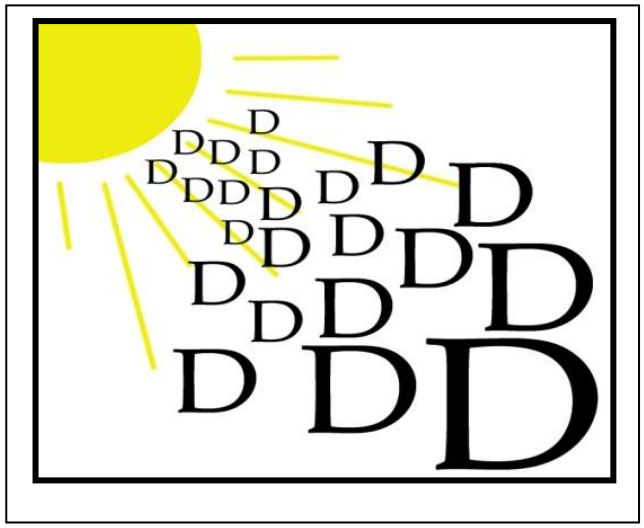
Supervisor:
Dr. Gail Rees
School of Biomedical
and Biological Sciences
University of Plymouth
Gail.rees@plymouth.ac.uk
Tel: 01752 584647

If you feel your complaint has not been resolved please contact the secretary to the Faculty of Science and Technology Human Ethics Committee.
Mrs Paula Simson
01752 584503

University of Plymouth

Relation between vitamin D, body fat and health of Middle Eastern and Caucasian people

Participant Information Leaflet



- Vitamin D is not only important for bone health, but it may also help prevent many diseases such as cancer, diabetes and heart disease
- If you are overweight or obese you are more at risk of vitamin D deficiency
- Middle Eastern People are especially at risk of vitamin D deficiency as shown by our previous research
- More than half of the UK population has insufficient levels of vitamin D

Therefore, we would like to invite you to take part in a study to improve your health and vitamin D levels by losing weight

Below are some questions people often ask about research and our answers:

Who has approved this project?
The Research Ethics Committee for the Faculty of Science and Technology at the University of Plymouth has reviewed the project and given its ethical approval.

Why have I been chosen to take part?
We have invited people who wish to have a healthier lifestyle and reduce their weight / body fat. Your participation is voluntary but we hope you can help.

If you have any further questions please do not hesitate to contact one of the project team listed on the back of this leaflet.

What would be involved in taking part?

You will be asked to give fasting blood samples to assess vitamin D levels at the start, middle and end of the study to find out the effect of season and body fat loss on your vitamin D status. This will be carried out by a trained phlebotomist. From this sample we will also measure hormones that affect appetite. After 3 months we will provide you with advice to reduce your usual calorie intake and increase your physical activity to encourage weight loss over a 3 month period. A qualified dietitian will meet with you once a month for 3 months to give you individual advice. You will be asked to keep a food diary for 3 days and physical activity will be estimated via a questionnaire. Also weight, height, waist circumference and hip circumference will be measured. Body fat will be measured using body fat scales. These measurements will be taken at the start and monthly after the first 3 months. We will also collect some information on sun exposure and medications in a questionnaire.

What if I change my mind?

You can withdraw from the project at any time without having to give an explanation.

Will taking part be of any benefit to me?
This study will give you information and advice on a healthy diet and physical activity for achieving a healthy body weight. This will be individualized for you. You will also be told about your vitamin D levels at the end of the study and whether you need to increase vitamin D rich foods or take supplements.

Are there disadvantages to taking part?
We recognise taking part will take up some of your time. We will do our best to minimise any inconvenience by ensuring that we meet at a time and place convenient for you. We do not expect anyone to suffer any harm or injury as a result of participating in this project. Blood collection may be uncomfortable or cause a small bruise.

Will my data be confidential?
You will receive your own code which will be the only link to you and this code will be stored separately on a computer that is secure and accessible only by the research team. The code will allow us to remove your data from the project if you change your mind after participating.

What if I have any concerns?
If you think of questions about the project please feel free to contact the research team using the contact details on the back of this leaflet.

How and where will the results be published?
We plan to publish our results in academic journals, present at scientific and nutrition meetings and conferences. We will also send you a summary of the research findings when the project is complete.

Thank you for reading this leaflet and for considering helping with this study

APPENDIX 7

Poster for Recruitment

Did you know:

- **Vitamin D is not only important for bone health, but it may also help prevent many diseases such as cancer, diabetes and heart disease**
- **If you are overweight you are more at risk of vitamin D deficiency**
- **Middle Eastern people are especially at risk of vitamin D deficiency as shown by our previous research**
- **More than half of the UK population has insufficient levels of vitamin D**

So, would you like to improve your health and vitamin D levels by losing weight?

If you answered **yes** to our question, come and find out more about our new research on vitamin D status and weight loss.

We are looking for Middle Eastern and Caucasian volunteers aged 18 years and over to take part.

You will receive *free* consultations with registered dietitians to achieve a healthy diet and lifestyle. While you are losing weight we will measure your vitamin D levels. At the end of the study you will have lost weight and know more about your vitamin D status.

For further information please contact by email:

Prof. Anne E de Looy adelooy@plymouth.ac.uk

Dr Gail Rees gail.rees@plymouth.ac.uk

Wassan Ahmed wassan.ahmed@plymouth.ac.uk

APPENDIX 8

Demographic Screening Questionnaire

Subject No. ☐

Demographic screening questionnaire

Personal information

Answer by ticking (FAO/WHO/UNU) below:

What is your age? 19-29 ☐

30-39 ☐

40-49 ☐

50-59 ☐

60+ ☐

What is your gender? Male ☐ Female ☐

What is your ethnic origin? Caucasian ☐ Middle Eastern ☐

Any other background, please write in ……

How long you have been in the UK? ……

Are you pregnant? Yes ( ) No ( ) N/A ( )

Are you breasting feeding? Yes ( ) No ( ) N/A ( )

Have you lost weight during the last 3 months? Yes ( ) No ( )

Do you smoke cigarettes? Yes ( ) No ( ) recently given-up ( )

Do you do outdoor activities with exposure to sunlight (for your job or as leisure time)?

Yes ( ) No ( )

If yes, how long do you spend outside in daylight each day? ……

Any days different? ……
……

Do you use sunscreen? Yes ( ) No ( )

If yes, what is the sun protection factor (SPF) number of the sunscreen you use most often?

2 ( ) 8 ( ) 15 ( ) 20 ( ) 30 ( ) 45 ( ) 60 ( )

Does your dress style often include?

Sunglasses ( ) Long sleeved shirt ( ) Headscarf ( ) Veil ( ) Hat ( )

What is your Occupation? ……..

Do you usually follow a special diet? e.g. vegetarian, cholesterol lowering, weight reducing?

Yes ( ) No ( )

If yes, Please specify ……

……

Medical information

(Please answer the following questions as accurately as possible)

List any prescribed or over the counter medications you take or have taken; including the contraceptive pill / H.R.T. for women (in last month):

…… ……
…… ……

Do you currently take a nutritional supplement (Vitamins/Minerals)?

Yes ( ) No ( ) If yes, please describe the supplements you took below:

…… ……
…… ……

Do you have pacemaker? Yes ( ) No ( )

Please, take a moment to fill in any questions that you may have skipped

Thank you very much for your co-operation and time ☺

APPENDIX 9

Physical Activity Questionnaire

INTERNATIONAL PHYSICAL ACTIVITY QUESTIONNAIRE

We are interested in finding out about the kinds of physical activities that people do as part of their everyday lives. The questions will ask you about the time you spent being physically active in the **last 7 days**. Please answer each question even if you do not consider yourself to be an active person. Please think about the activities you do at work, as part of your house and yard work, to get from place to place, and in your spare time for recreation, exercise or sport.

Think about all the **vigorous** activities that you did in the **last 7 days**. **Vigorous** physical activities refer to activities that take hard physical effort and make you breathe much harder than normal. Think *only* about those physical activities that you did for at least 10 minutes at a time.

During the **last 7 days**, on how many days did you do **vigorous** physical activities like heavy lifting, digging, aerobics, or fast bicycling?

_____ **days per week**

☐ No vigorous physical activities → ***Skip to question 3***

How much time did you usually spend doing **vigorous** physical activities on one of those days?

_____ **hours per day**
_____ **minutes per day**

☐ Don't know/Not sure

Think about all the **moderate** activities that you did in the **last 7 days**. **Moderate** activities refer to activities that take moderate physical effort and make you breathe somewhat harder than normal. Think only about those physical activities that you did for at least 10 minutes at a time.

During the **last 7 days**, on how many days did you do **moderate** physical activities like carrying light loads, bicycling at a regular pace, or doubles tennis? Do not include walking.

_____ **days per week**

☐ No moderate physical activities → ***Skip to question 5***

How much time did you usually spend doing **moderate** physical activities on one of those days?

_____ **hours per day**
_____ **minutes per day**

☐ Don’t know/Not sure

Think about the time you spent **walking** in the **last 7 days**. This includes at work and at home, walking to travel from place to place, and any other walking that you might do solely for recreation, sport, exercise, or leisure.

5. During the **last 7 days**, on how many days did you **walk** for at least 10 minutes at a time?

_____ **days per week**

☐ No walking → ***Skip to question 7***

How much time did you usually spend **walking** on one of those days?

_____ **hours per day**
_____ **minutes per day**

☐ Don’t know/Not sure

The last question is about the time you spent **sitting** on weekdays during the **last 7 days**. Include time spent at work, at home, while doing course work and during leisure time. This may include time spent sitting at a desk, visiting friends, reading, or sitting or lying down to watch television.

During the **last 7 days**, how much time did you spend **sitting** on a **week day**?

_____ **hours per day**
_____ **minutes per day**

☐ Don’t know/Not sure

This is the end of the questionnaire, thank you for participating.

APPENDIX 10

Food and Drink Diary Instructions

Food and Drink Diary Instructions

Please read though these pages before starting your diary

We would like you to keep this diary of everything you eat and drink over 3 days. Please include all food consumed at home and outside the home e.g. work, college or restaurants. It is very important that you do not change what you normally eat or drink just because you are keeping this record. Please keep to your usual food habits.

Day and date

Please write down the day and date at the top of the page each time you start a new day of recording.

Time slots

Please note the time of each eating occasion into the space provided. For easy use each day is divided into sections, from the first thing in the morning to late evening and through the night.

What do you eat?

Please describe the food you eat in as much detail as possible. Page14-19 will help with sort of detail we need. Like **cooking methods** (fried, grilled, baked etc.) and any **additions** (fats, sugar/sweeteners, sauces, pepper etc.).

- **Homemade dishes:** if you have eaten any **homemade dishes** e.g. chicken casserole, please record the name of the recipe, ingredients with amounts (including water or other fluids) for the whole recipe, the number of people the recipe serves, and the cooking method. Write this down in the recipe section at the end of the record day. Record how much of the whole recipe you have eaten in the portion size column (see examples on pages 4-13).
- **Take –aways and eating out:** If you have eaten **Take –aways** or made up dishes not prepared at home such as at restaurant or a friend's house, please record as much detail about the ingredients as you can e.g. vegetable curry containing chickpeas, aubergine, onion and tomato.

Brand name

Please note the **brand name** (if known). Most packed foods will list a brand name, e.g. Bird's eye, Hovis, or supermarket own brands.

- **Labels/Wrappers:** labels are an important source of information for us. It helps us a great deal if you enclose, in the plastic bag provided, labels from all **ready meals,** labels from **foods of lesser known brands**.

Portion sizes

Examples for how to describe the **quantity** or **portion size** you had of a particular food or drink are shown on pages 14-19.

For foods, quantity can be described using:

- **Household measures,** e.g. one teaspoon (tsp) of sugar, two thick slices of bread, 4 tablespoons (tbsp) of peas, ½ cup of gravy. Be careful when describing amounts in spoons that you are referring to the correct spoon size. Compare the spoons you use with the life size pictures at the back of this diary.
- **Weights from labels,** e.g. 4 oz steak, 420g tin of baked beans, 125g pot of yoghurt.
- **Number of items,** e.g. 4 fish fingers, 2 chicken nuggets, 1 Rich Tea biscuit
- **Picture examples** for specific foods on pages 20-23.

For drinks, quantity can be described using:

- The **size of glass, cup etc** (e.g. large glass) or the **volume (**e.g. 300ml). Examples of typical drinks containers are on pages 24-25.
- **Volumes from labels** (e.g. 330ml can of fizzy drink).

We would like to know the **amount that was actually eaten** which means taking into account leftovers. You can do this in two ways:

1- Record what was served and note what was not eaten e.g. 3 tbsp of peas, only 2 tbsp eaten; 1 weetabix, ate ½.
2- Only record the amount actually eaten i.e. 2 tbsp of peas; ½ weetabix.

When to fill in the diary

Please record your eating as you go, not from memory at the end of the day. Use written notes on a pad if you forget to take your diary with you. Each diary day covers a 24 hr period, so please include any food or drinks that you may have had during the night. Remember to include foods and drinks between meals (snacks) including water.

- **Overleaf you can see 2 example days that have been filled in by different people. These examples show you how we would like you to record your food and drink, for example a ready meal and a homemade dish. Your instruction booklet contains further examples such as how to describe food eaten in a restaurant.**
- **If you have any queries about how to complete the diary please contact the researcher.**

It only takes a few minutes for each eating occasion!
For your convenience a separate booklet with instructions and examples is provided.

All the information you give us is strictly confidential. It will only be used for research purposes. Only your subject number appears on the record form. Nobody will be able to identify you from the record form.

• Thank you for your co-operation and time - we really appreciate it! 

Middle East sample

Day *Thurs*	**Date** *31 st March*		
Time	**Food/Drink description preparation**	**Brand name**	**Portion size or quantity eaten**
	6 am to 9 am		
8 am	*Tea* *Milk (fresh, semi-skimmed)* *Sugar white* *Toast, white medium sliced* *Blue cheese* *Jam* *Fried egg* *Tomato* *Olive oil* *Salt* *Black pepper*	*Twinings* *Silverspoon* *Kingsmill* *Rosenborg Danish* *Hartleys* *Sainsbury's basics* *Sainsbury's (loose)* *Bertolli Extra Virgin* *Saxa Table Salt* *Schwartz*	*Mug* *A little* *1 level tsp* *3 slice* *10 g* *1 heaped tsp* *1* *20 g* *2 tablespoons* *A little* *A little*
	9 am to 12 noon		

Time	Food/Drink description preparation	Brand name	Portion size or quantity eaten
	12 noon to 2 pm		
1.30 pm	*Grilled fish* *Salt* *Bread, White Pitta* *Pickles(Chutney)* Mixed salad (Iceberg lettuce, tomato, spring onion, olive oil, salt, wine vinegar, 1 half of a squeezed lemon)	*Carp* *Saxa Table Salt* *Sainsbury's ow* Geeta's Lime & Chilli	*120 g* *A little* *1 large* *1 level tsp* *1 bowl*
	2 pm to 5 pm		
	5 pm to 8 pm		
5.30 pm	*Black tea* *Sugar white* *Cookies stuffed with dates (Ma`amoul) (see recipe)*	*Twinings* *Silverspoon* *Homemade*	*Large cup* *1 level tsp* *2 pieces(25g each one)*

Time	Food/Drink description preparation	Brand name	Portion size or quantity eaten
	8 pm to 10 pm		
8.30 pm	*Potato crisps, Ready Salted* *Mixed nuts* *Banana*	*Walkers*	*25 g bag from multipack* *50 g* *Medium size*
	10 pm to 6 am		
10.30 pm	*Lentil, Tomato & Vegetable Soup* *Red wine*	*Sainsbury's own* *Australian Merlot*	*200g* *1 unit*

Did you **finish all the food and drink** that you recorded in the diary today?

Yes ☐ No ☐

If no, please **go back to the diary and make a note of any leftover**

Please record over the page details of any recipes or (if not already described) ingredients of made up dishes or take-away dishes.

Write in recipes or ingredients of made up dishes or take-away dishes

NAME OF DISH *Cookies stuffed with dates (Ma`amoul)* ***Yield:*** *20 pieces*

Ingredients	***Amount***	***Ingredients***	***Amount***
Active dry yeast	*½ teaspoon*		*Dash*
Baking powder	*1 teaspoon*		
Warm water	*¼ cup*		
ground cardamom	*1 teaspoon*		
nutmeg	*¼ teaspoon*		
Egg	*1 large*		
Margarine	*½ cup*		
All-purpose flour	*2 cup*		
milk	*¼ cup*		
Sugar	*2 tablespoon*		
salt	*¼ teaspoon*		
pitted soft dates	*¾ cup*		

Brief description of cooking method

Dissolve the yeast in the water and add the flour, egg, milk, margarine, sugar, salt, ground cardamom, nutmeg and baking powder. Mixing till the dough hold together. Roll into 20 equally sized balls.

Place the date in the centre of each round and draw the edges up and around the filling.

Bake for 25 to 30 min.

Day *Friday*	**Date** *28.09.2007*		
Time	**Food/Drink description preparation**	**Brand name**	**Portion size or quantity eaten**
	6 am to 9 am		
7 am	*Black tea* *Sugar white* *Cheese* *Toast, Wholemeal Medium sliced*	*Tetley* *Fairtrade Granulated* *Mozzarella Maxi* *Hovis*	*Mug* *1 level tsp* *50g* *1 slice*
	9am to 12 noon		
11 am	*Green apple* *Orange* *Banana* *Water*		*1, medium size* *1, medium size* *1, medium size* *300 ml glass*
	12 noon to 2 pm		
12 pm	*Milk chocolate*	*Kit Kat*	*2 Finger*
	2pm to 5pm		
2 pm	*Fish & chips* *Soft drink*	 *7 up*	*1 Large portion* *330 ml can*
	5 pm to 8 pm		
5 pm *7.30 pm*	*Orange juice* *Ice cream*	*Tropicana* *Magnum Classic*	*300 ml glass* *360 ml, 1 stick*

Time	Food/Drink description preparation	Brand name	Portion size or quantity eaten
	8pm to 10pm		
9.15 pm	*Aubergine sauce with Lamb meat (see recipe)* *White rice* *pickled vegetables* Mixed salad (Iceberg lettuce, tomato, cucumber, green pepper) *Soft drink*	 *Sprite*	*6 tablespoons* *(100 g lamb meat)* *100 g* *50 g* *100g* *330 ml can*
	10pm to 6am		
10.30 pm	*dates* *Watermelon*		*15, large size* *100g*

Did you **finish all the food and drink** that you recorded in the diary today?

Yes ☐ No ☐

If no, please **go back to the diary and make a note of any leftover**

Please record over the page details of any recipes or (if not already described) ingredients of made up dishes or take-away dishes

Write in recipes or ingredients of made up dishes or take-away dishes

NAME OF DISH *Aubergine sauce* ***SERVES:*** *3*

Ingredients	***Amount***	***Ingredients***	***Amount***
sunflower oil	*1 tablespoon*		*Dash*
Chopped Lamb meat	*350 g*		
Aubergine, peeled and chopped	*1 large*		
Tomato, chopped	*1 large*		
onion, chopped	*1 large*		
Green peppers, chopped	*1medium*		
Potato, peeled and chopped	*1 medium*		
garlic	*5 cloves*		
Tomato puree	*½ tablespoon*		
water	*60 ml*		
Allspice	*1 teaspoon*		
Salt	*1 teaspoon*		
Black pepper	*1 teaspoon*		

Brief description of cooking method

Fry the meat& mix in the aubergine, tomato, onion, green pepper, potato and garlic. Cook and stir until tender and lightly browned.

Blend the tomato puree and water. Mix with the lamb. Season with allspice, salt and pepper. Simmer about 11/2 hours.

Caucasians sample

Day *Thurs*	**Date** *31 st March*		
Time	**Food/Drink description preparation**	**Brand name**	**Portion size or quantity eaten**
	6 am to 9 am		
6.30 am	*Filter coffee, decaffeinated* *Milk (fresh, semi-skimmed)* *Sugar white*	*Douwe Egberts*	*Mug* *A little* *1 level tsp*
7.30 am	*Filter coffee with milk and sugar* *Cornflakes* *Milk (fresh, semi-skimmed)* *Toast, granary medium sliced* *Light spread* *marmalade*	*As above* *Tesco's own* *Hovis* *Flora* *Hartleys*	*As above* *1b* *Drowned* *1 slice* *Med spread* *1 heaped tsp*
	9 am to 12 noon		
10.15 am	Instant coffee, not *decaffeinated* *Milk (fresh, whole)* *Sugar brown*	*Kenco*	*Mug* *A little* *1 level tsp*
11 am	*Digestive biscuit – chocolate coated on one side*	*McVities*	*2*

Time	Food/Drink description preparation	Brand name	Portion size or quantity eaten
	12 noon to 2 pm		
12.30 pm	*Ham salad sandwich from home* *Bread, wholemeal, thick sliced* *Light spread* *Low fat Mayonnaise* *Smoked ham thinly sliced* *Lettuce, iceberg* *Cucumber with skin* *Unsweetened orange juice from canteen* *Apple with skin from home, Braeburn*	 *Tesco's own* *Flora* *Hellmans* *Tesco's own* *Tropicana*	 *2 slices* *Thin spread on 1slice* *2 teaspoons* *2 slices* *1 leaf* *4 thin slices* *250 ml carton* *Medium size, core left*
	2 pm to 5 pm		
3 pm	*Tea, decaffeinated* *Milk (fresh, whole)* *Jaffa cake – mini variety*	*Twinings* *Tesco's own* *McVities*	*Mug* *Some* *6*

Time	Food/Drink description preparation	Brand name	Portion size or quantity eaten
	5 pm to 8 pm		
6.30 pm	*Gin* *Tonic water diet* *Lager* *Salted peanuts*	*Gordon's* *Schweppes* *Draught* *KP*	*Single measure* *½ small glass* *1 pint* *1 handful*
8 pm	*Spaghetti, wholemeal* *Bolognese sauce (see recipe)* *Courgettes (fried in butter)* *Tinned peaches in juice 9juice drained)* *Single cream UHT* *Orange squash No Added Sugar*	*Tesco's own* *Prince's* *Sainsbury's own*	*3b* *6 tablespoons* *4 tablespoons* *3 halves* *1 tablespoons* *200 ml glass, 1 part squash, 3 parts tap water*
	8 pm to 10 pm		
9 pm	*Grapes, green, seedless* *Chocolates, chocolate creams* *Potato crisps, Prawn cocktail*	 *Bendicks* *Walkers*	*15* *2* *25 g bag from multipack*
	10 pm to 6 am		
10.30 pm	*Camomile tea* *(no milk or sugar)*	*Twinings*	*1 mug*

Did you **finish all the food and drink** that you recorded in the diary today?

Yes ☐ No ☐

If no, please **go back to the diary and make a note of any leftover**

Please record over the page details of any recipes or (if not already described) ingredients of made up dishes or take-away dishes.

Write in recipes or ingredients of made up dishes or take-away dishes

NAME OF DISH *Bolognese sauce* ***SERVES:*** *4*

Ingredients	***Amount***	***Ingredients***	***Amount***
Co-op low fat beef mince	*500g*	*Lea & Perrins Worcester sauce*	*Dash*
garlic	*3 cloves*		
onion	*1 medium*		
sweet red pepper	*1 medium*		
Napoli chopped tomatoes	*400g tin*		
Tesco tomato puree	*1 tablespoon*		
Tesco olive oil	*1 tablespoon*		
mixed herbs	*1 dessertspoon*		

Brief description of cooking method

Fry onion & garlic in oil, add mince and fry till brown.

Add pepper, tomatoes, puree, Worcester sauce & herbs. Simmer for 30 mins.

Day *Friday*	**Date** *28.09.2007*		
Time	**Food/Drink description preparation**	**Brand name**	**Portion size or quantity eaten**
	6 am to 9 am		
8.00 am *8.45 am*	*Cappuccino, no sugar* *Blueberry muffin, regular not low fat* *Tap water*	*Starbucks* *Starbucks*	*Medium size* *One* *300 ml glass*
	9am to 12 noon		
10 am	*Banana* *Black tea* *semi-skimmed milk, no sugar*	 *Typhoo* *Asda*	*One, medium size* *Large Mug* *A lot*
	12 noon to 2 pm		
1 pm	*Crayfish sandwich* *multiseed bread, wholemeal, medium cut,* *crayfish in lemon mayonnaise, no other spread* *rocket leaves* *Apple & Raspberry fruit drink*	*M&S pre-packed Sandwich* *J2O*	 *2 slices* *Medium filling* *6 to 8* *1 bottle, 275ml*

Time	Food/Drink description preparation	Brand name	Portion size or quantity eaten
	2pm to 5pm		
4.30 pm	*Coffee, instant* *Semi-skimmed milk* *Fairy Cake, homemade, see recipe*	*Kenco*	*Medium mug* *A lot* *1 cake*
	5pm to 8pm		
7.30 pm	*Chicken in creamy mushroom and white wine sauce for 2, oven* *White rice, boiled* *Wine*	*Sainsbury's, 370g (wrapper collected)* *Easy cook, Italian, Sainsbury's* *Sauvignon Blanc New Zealand*	*½ pack* *2C* *1 small glass, 125ml*
	8pm to 10pm		
9.15 pm	*Squash, apple & blackcurrant, no added sugar* *Crisps*	*Sainsbury's* *Pringles, sour cream and chives*	*1 average glass, 200ml* *5*
	10pm to 6am		
11.30 pm	*Water*	*tap*	*1 medium glass*

Did you **finish all the food and drink** that you recorded in the diary today?

Yes ☐ No ☐

If no, please **go back to the diary and make a note of any leftover**

Please record over the page details of any recipes or (if not already described) ingredients of made up dishes or take-away dishes

Write in recipes or ingredients of made up dishes or take-away dishes

NAME OF DISH *Fairy Cakes* ***SERVES:*** *makes 20 cakes*

Ingredients	***Amount***	***Ingredients***	***Amount***
Tate & Lyle caster sugar	*175g*	*Silver Spoon icing sugar*	*140g*
Anchor butter, unsalted	*175g*	*Yellow food colouring*	*3 drops*
Co-op eggs	*3*	*Water*	*2 tablespoons*
Homepride self-raising flour	*175g*		
Baking powder	*1 teaspoon*		

Brief description of cooking method

Mix together and bake for 15 min.

Mix icing sugar with water and add colouring. Approx. 1 teaspoon of icing on each cake.

Food/Drink	***Description & Preparation***	***Portion size or quantity***
Bacon	Back, middle, streaky; smoked or un-smoked; fat eaten; dry-fried or fried in oil/fat (type used) or grilled rashers	Number of rashers
Baked beans	Standard, reduced salt or reduced sugar	Tablespoons, weight of beans marked on tin label (e.g. 420g)
Beefburger (hamburger)	Home-made (ingredients), from a packet (brand name) or take-away; fried (type of oil/fat), microwaved or grilled; economy; with or without bread roll	Number, large or small, ounces or in grams if info on package
Beer	What sort e.g. stout, bitter, lager; draught, canned, bottled; low-alcohol or home-made	Number of pints or half pints, size of can or bottle
Biscuits	What sort and brand e.g. cheese, wafer, crispbread, sweet, chocolate, shortbread, home-made	Number, size (standard or mini variety)
Bread (see also sandwiches)	Wholemeal, granary, white or brown; currant, fruit, malt; large or small loaf; sliced or unsliced loaf; give brand	Number of slices; thick, medium or thin slices
Bread rolls	Wholemeal, white or brown; alone or with filling; crusty or soft	Size, number of rolls
Breakfast cereal (see also porridge)	What sort and brand e.g. Kellogg's cornflakes; any added fruit and/or nuts; Muesli – added sugar and/or fruit	Tablespoons or picture 1
Bun	Iced, currant or plain, homemade or bought (brand name)	Large or small, number
Butter, margarine & fat spreads	Give full product name	Thick/average/thin- spread; spoons

Food/Drink	***Description & Preparation***	***Portion size or quantity***
Cake	Individual or piece of large; type and brand; fruit (rich), sponge, fresh cream, buttercream, iced; type of filling	Number, slices, packet weight, see picture 10 for sponge cake
Cheese	Name, brand and type e.g. cheddar, cream, cottage, soft; low fat	picture 9, or number of slices, thick or thin cut, number of spoons
Chips	Fresh, frozen, oven, microwave, take-away (where from); thick/straight/crinkle/fine cut; type of oil/fat used for cooking give brand name	picture 4, as A, B, or C or 2 x B, etc
Chocolate(s)	What sort e.g. plain, milk, white, fancy, diabetic; type of filling; give brand name	Number, weight/size of bar
Coffee	With milk (see section on milk); half milk/half water; all milk; ground/filter, instant; decaffeinated; give brand name	Cups or mugs
Cream	Single, whipped, double or clotted; dairy or non-dairy; low-fat; fresh, UHT/Long-life; imitation cream e.g. Elmlea	Tablespoons
Crisps	What sort e.g. potato, corn, wheat, maize, vegetable etc; give brand; flavour; low-fat or low-salt; premium variety e.g. Kettle chips, Walker's Sensations	Packet weight
Custard	Pouring custard or egg custard; made with powder and milk/sugar, instant, ready to serve (tinned or carton); low fat, sugar free, brand	Tablespoons
Doughnut	Plain, jam, cream or iced; round or ring, where bought/brand name	Number, size e.g. mini, large
Egg	Boiled, fried (type of oil/fat), scrambled (type of fat used, with or without added milk), poached, omelette (with or without filling, type of oil/fat used), etc	Number of eggs, large, medium or small eggs
Fish (including canned)	What sort and brand e.g. cod, tuna; fried (type of oil/fat), grilled, poached (water or milk) or steamed; with batter or breadcrumbs; canned in oil, brine or tomato sauce	Size of can or spoons (for canned fish) or picture 7 for battered fish

Food/Drink	***Description & Preparation***	***Portion size or quantity***
Fish cakes & fish fingers	Type of fish; plain or battered or in breadcrumbs; fried, grilled, baked or microwaved; economy	Size, number packet weight
Fruit - fresh	What sort; eaten with or without skin	Size, number
Fruit - stewed/canned	What sort; sweetened or unsweetened; in fruit juice or syrup; juice or syrup eaten	Tablespoons Size of can or weight on can
Fruit – juice (pure)	What sort and brand e.g. apple, orange; sweetened or unsweetened; pasteurised or UHT/Longlife; freshly squeezed; added vitamins/minerals, omega 3?	Glass (size or volume) or carton size
Ice cream	Flavour; dairy or non-dairy; brand name; luxury/premium; added nuts, fruit	Number of tablespoons/ scoops
Jam, honey	What sort; low-sugar/diabetic; shop bought/brand or homemade	Teaspoons, heaped or level, or thin or thick spread
Marmalade	Type and brand; low-sugar; thick cut; shop bought/brand or homemade	Teaspoons, heaped or level, or thin or thick spread
Meat (see also bacon, burgers & sausages)	What sort; cut of meat e.g. chop, breast, minced; lean or fatty; fat removed or eaten; skin removed or eaten; how cooked; with or without gravy	Large/small/medium, tablespoons, or picture 6 for stew portion
Milk	Brand and type (whole, semi-skimmed, skimmed); fresh, sterilized, UHT, dried; soya milk (sweetened/unsweetened), goats' milk, rice milk; flavoured; fortified with added vitamins and/or minerals	Pints, glass (size or volume) or cup. For milk on cereal: *Damp/normal/drowned.* For milk in tea/coffee: *a little/ some/ little/some/a lot*

Food/Drink	*Description & Preparation*	*Portion size or quantity*
Nuts	What sort and brand; dry roasted, ordinary salted, honey roasted; unsalted	Packet weight, handful
Pie (sweet or savoury)	What sort and brand; individual or helping; one pastry crust or two; type of pastry	Individual or slice, or picture 8
Pizza	Thin base or deep pan or French bread; topping; brand name and type	Individual, slice, fraction of large pizza e.g. ¼
Porridge	Brand name; made with oats or cornmeal or instant oat cereal; made with milk and/or water; with sugar or honey; with milk or cream	Bowl
Potatoes (see also chips)	Old or new; baked, boiled, roast (type of oil/fat); skin eaten; mashed (with butter/spread and with or without milk); fried/chips (type of oil/fat); instant; any additions e.g. butter	Mash – tablespoons, number of half or whole potatoes, small or large potatoes, or picture 4 for chips portion
Pudding	What sort; e.g. steamed sponge; with fruit; mousse; instant desserts; milk puddings	Tablespoons, picture 10 for slice of sponge
Rice	What sort; e.g. basmati, easy cook, long or short grain; white or brown; boiled or fried (type of oil/fat); brand name	Tablespoons or picture 2
salad	Ingredients; if with dressing what sort (oil and vinegar, mayonnaise); brand name of dressing	Amount of each component; e.g. number of tomatoes, slices of cucumber, leaves; tablespoons of dressing

Food/Drink	***Description & Preparation***	***Portion size or quantity***
Sandwiches and rolls	Type of bread/roll (see Bread & Rolls); butter or margarine; type of filling; including salad, mayonnaise, pickle etc. If shop-bought, where from?	Number of rolls or slices of bread; amount of butter/margarine (on both slices?); amount of filling
Sauce – cold (including mayonnaise)	Tomato ketchup, brown sauce, soy sauce, salad cream, mayonnaise; low fat; brand name	Teaspoons, tablespoons
Sausages	What sort; e.g. beef, pork; fried (type of oil/fat) or grilled; low fat; economy; brand name	Large or small, number
Sausage rolls	Type of pastry; brand name	Number, size e.g. jumbo, Standard, mini
Scone	Fruit, sweet, plain, cheese; type of flour; bought/brand or homemade	Number, small, medium r large
Savoury snacks – in packet	What sort: e.g. Cheddars, cheese straws, Twiglets, Pretzels; give, brand name	Size (standard or mini variety), packet weight, number
Soft drinks – squash/ concentrate/cordial	Give brand name & flavour; no added sugar/low calorie/sugar free; “high” juice; fortified with added vitamins and/or minerals	Glass (size or volume)
Soft drinks – carbonated/fizzy	Give brand & flavour; diet/low-calorie; canned or bottled; cola – caffeine free	Glass, can or bottle (size or volume)
Soft drinks – ready to drink	Give brand & flavour; no added sugar/low calorie/sugar free; does it contain real fruit juice, if so, how much?; fortified with added vitamins and/or minerals	Glass, carton or bottle (size or volume)
Soup	What sort; give brand name; cream or clear; canned, packet, instant or vending machine, home-made	Tablespoons, bowl or mug
Spaghetti, other pasta	What sort; fresh/chilled or dried; white, wholemeal; canned in sauce; type of filling if ravioli, cannelloni etc	Tablespoons (or how much dry pasta used per portion in grams/packet size) or picture 3

Food/Drink	***Description & Preparation***	***Portion size or quantity***
Spirits	What sort: e.g. whisky, gin, vodka, rum	Measures as in pub
Sugar	Added to cereals, tea, coffee, fruit, etc; what sort; e.g. white, brown, demerara	Heaped or level Teaspoons
Sweets	What sort: e.g. toffees, boiled sweets, diabetic; give brand name	Number, packet weight
Tea	With/without milk (see section on milk); decaffeinated, herb	Mugs or cups
Vegetables (not including potatoes)	What sort; how cooked or raw; additions e.g. butter, other fat or sauce	Tablespoons, number of florets or sprouts, weight from tins or packet as guidance
Water	Tap, filtered, bottled: give brand name	Glass or bottle (size or volume)
Wine, sherry, port	White, red; sweet, dry; low-alcohol; give brand name	Glass (size or volume)
Yoghurt, fromage frais	What sort: e.g. natural/plain or flavoured; creamy, Greek, low-fat, very low fat/diet, soya; with fruit pieces or just fruit flavoured; twinpot with separate cereal/crumble; fortified with added vitamins and/or minerals; brand name	Pot size or tablespoons
Home-made dishes	Please say what the dish is called (record recipe or details of dish if you can in the section provided) and how many persons it serves	Tablespoons – heaped or level, number, size
Ready-made meals	Please give brand name and full description of product; did it contain any accompaniments e.g. rice, vegetables, sauces; was it chilled or frozen; microwaved, oven cooked, boil-in-the-bag; was it low fat, healthy eating range. Enclose label and ingredients list if possible in your plastic bag	Packet weight, if not whole packet describe portion consumed
Take-away food or food eaten out	Please say what the dish is called and give main ingredients if you can. Give name of a chain restaurant e.g. McDonalds	Tablespoons, portion size e.g. small/medium/large

Use the pictures to help you indicate the size of the portion you have eaten.
Write on the food record the picture number and size A, B or C nearest to your own helping.

Remember that the pictures are much smaller than life size.
The actual size of the dinner plate is 10 inches (25cm), the side plate, 7 inches (18cm), and the bowl, 6.3 inches (16cm).

The tables on pages 16-21 also give examples of foods that you might eat and how much information is required about them.

1. Breakfast cereals

2. Rice

3. Spaghetti

4. Chips

5. Broccoli or cauliflower

6. Stew or curry

7. Battered fish

8. Quiche / Pie

9. Cheese

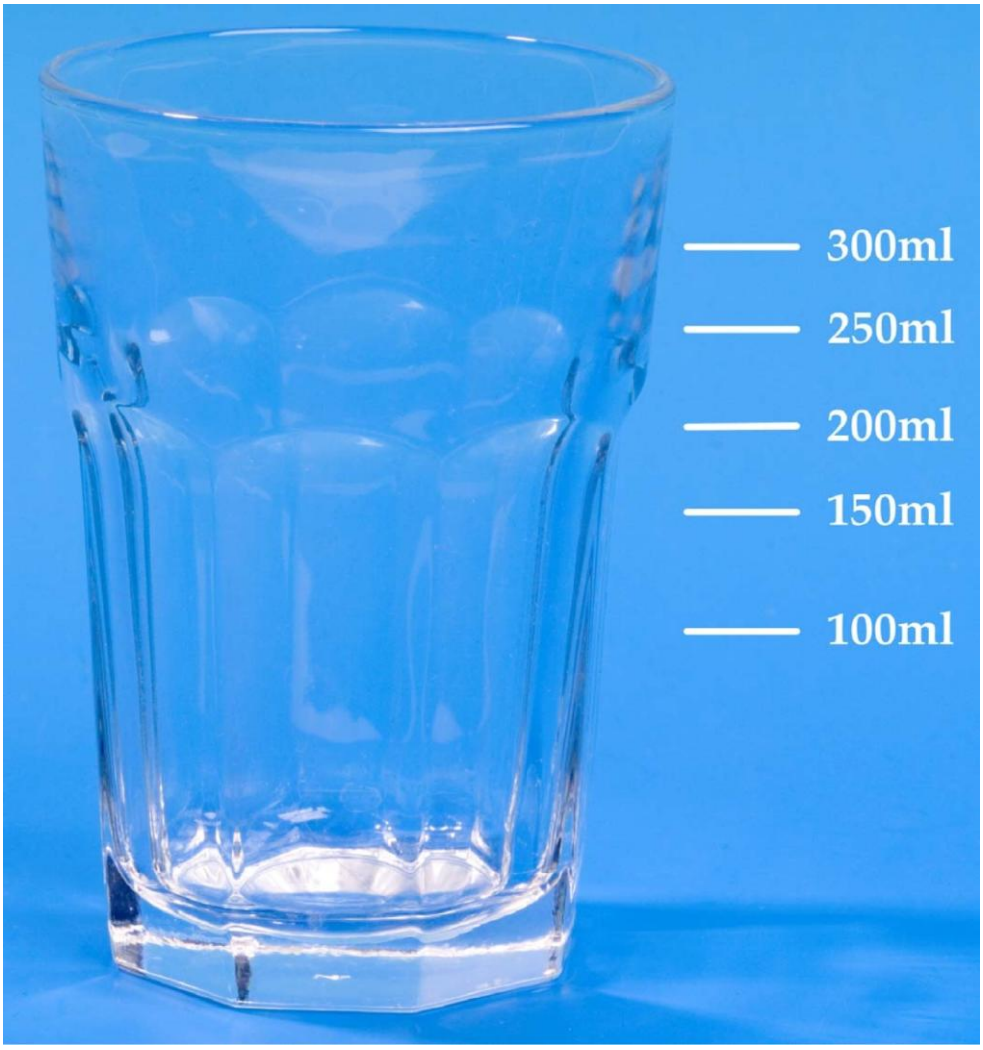
10. Sponge cake

Typical quantities of drinks in various containers measured in millilitres (ml)

	Small Glass	Average Glass	Large Glass	Vending Cup	Cup	Mug
Soft Drinks	150	200	300			
Hot Drinks				170	190	260

Here is a life size glass showing what typical quantities look like. You can use this picture as a guide for estimating how much volume of drink the glass holds you are drinking from.

Life Size Glass



APPENDIX 11

Food Diary Form

Food diary form

Summary Sheet

First of all, we wish to thank you for giving up your time to fill in this form and we are very grateful for the help and support you are giving this study.

- The information on this form will help us to understand your dietary habits and nutritional status so we can assess your vitamin D intake, and at the next stage of the study give you an advice for weight loss.
- All the instructions you need are given (**Please Read the Instructions carefully).**
- Two examples of how to record 2 days food intake are also given.
- You should eat exactly as you normally do.
- I would like to ask you to record details of foods and beverages at the time they are consumed for 3 days.

- It is very important that the information you give us as accurate as possible. If you are having any doubts or difficulties do not hesitate to contact me for help.
- I will contact you in one week to collect the diet diary.
- We will meet again on …….. at ……in the nutrition lab (Link Building) for blood collection.

- It is really important that you come fasted and have not eaten or drunk anything other than water for 12 hours prior to this time. After blood collection we will provide you with a drink and a snack.
- After this we meet again in 3 months time to recheck your vitamin D status, appetite hormones, diet and exercise patterns and give you diet and lifestyle advice. I will contact you nearer the time to arrange the appointment.

Telephone: **Wassan Ahmed 01752 584695**

Email: Wassan.Ahmed@plymouth.ac.uk

ALL THE INFORMATION RECEIVED IN THIS SURVEY IS CONFIDENTIAL

THANK YOU FOR YOUR COOPERATION ☺

Food diary form

Day 1	**Date**		
Time	**Food/Drink description preparation**	**Brand name**	**Portion size or quantity eaten**
6 am to 9 am			
9 am to 12 noon			
12 noon to 2 pm			
2 pm to 5 pm			

Time	Food/Drink description preparation	Brand name	Portion size or quantity eaten
5 pm to 8 pm			
8 pm to 10 pm			
10 pm to 6 am			

Did you **finish all the food and drink** that you recorded in the diary today?

Yes ☐ No ☐

If no, please **go back to the diary and make a note of any leftover**

Please record over the page details of any recipes or (if not already described) ingredients of made up dishes or take-away dishes.

Write in recipes or ingredients of made up dishes or take-away dishe			
NAME OF DISH		***SERVES:***	
Ingredients	***Amount***	***Ingredients***	***Amount***
Brief description of cooking method			

Day 2	**Date**		
Time	**Food/Drink description preparation**	**Brand name**	**Portion size or quantity eaten**
6 am to 9 am			
9 am to 12 noon			
12 noon to 2 pm			
2 pm to 5 pm			

Time	Food/Drink description preparation	Brand name	Portion size or quantity eaten
5 pm to 8 pm			
8 pm to 10 pm			
10 pm to 6 am			

Did you **finish all the food and drink** that you recorded in the diary today?

Yes ☐ No ☐

If no, please **go back to the diary and make a note of any leftover**

Please record over the page details of any recipes or (if not already described) ingredients of made up dishes or take-away dish

Write in recipes or ingredients of made up dishes or take-away dishes			
NAME OF DISH		***SERVES:***	
Ingredients	***Amount***	***Ingredients***	***Amount***
Brief description of cooking method			

Day 3	**Date**		
Time	**Food/Drink description preparation**	**Brand name**	**Portion size or quantity eaten**
6 am to 9 am			
9 am to 12 noon			
12 noon to 2 pm			
2 pm to 5 pm			

Time	Food/Drink description preparation	Brand name	Portion size or quantity eaten
5 pm to 8 pm			
8 pm to 10 pm			
10 pm to 6 am			

Did you **finish all the food and drink** that you recorded in the diary today?

Yes ☐ No ☐

If no, please **go back to the diary and make a note of any leftover**

Please record over the page details of any recipes or (if not already described) ingredients of made up dishes or take-away dishes.

Write in recipes or ingredients of made up dishes or take-away dishes			
NAME OF DISH		***SERVES:***	
Ingredients	***Amount***	***Ingredients***	***Amount***
Brief description of cooking method			

General questions about your food/ drink in the last 3 days.

Milk

1- Which type of milk did you use most often in the last 3 days?

Whole, fresh, Pasteurised ☐

Semi-skimmed fresh, Pasteurised ☐

Skimmed (fat free) fresh, Pasteurised ☐

Dried ☐ *Type* _____

Soya ☐ *Type* _____

Other ☐ *Type* _____

Did not use ☐

Tea and coffee

2- How much milk did you usually have in coffee/ tea?

Coffee	A lot	☐	Some	☐	A little	☐	None/did not drink	☐
Tea	A lot	☐	Some	☐	A little	☐	None/did not drink	☐

3- Did you usually sweeten your coffee/ tea with sugar?

Coffee	Yes	☐	How many teaspoons in a mug/cup?	☐	No/did not drink	☐
Tea	Yes	☐	How many teaspoons in a mug/cup?	☐	No/did not drink	☐

4- Did you usually sweeten your coffee/ tea with artificial sweetener?

Coffee	Yes	☐	How many teaspoons in a mug/cup?	☐	No/did not drink	☐
Tea	Yes	☐	How many teaspoons in a mug/cup?	☐	No/did not drink	☐

Breakfast cereals

5- How much milk did you usually have on breakfast cereal?

Drowned ☐ Average ☐ Damp ☐ None/did nor eat ☐

Fats for spreading and cooking

6- Which type of fat spread did you use most often in the last 3 days?

Please record the full product name and fat content. _____

e.g. *Flora Omega 3 plus, low fat spread, 38% fat, polyunsaturated* _____

7- How thickly did you spread butter, margarine on bread, crackers etc?

Thick ☐ Medium ☐ Thin ☐ None ☐

8- Which type of cooking fat/oil did your household use

most often in the last 3 days? Please record the full product name.

e.g. Sainsbury's sunflower oil

Meat

9- If you ate meat in the last 3 days, what did you do with the visible fat?

Ate all ☐ Ate most ☐ Ate some ☐ Ate none of the fat ☐ Did not eat ☐

10- If you ate poultry in the last 3 days, did you eat the skin?

Always ☐ Sometimes ☐ Never ☐ Did not eat ☐

Thank you for completing this diary ☺

Subject No. ☐

APPENDIX 12

Data Collection Sheet

Data collection sheet

Subject No. ☐

Time / Measurement	**October 2010** Week 1/Baseline Date……. Time…….	**January 2011** Week 12 Date……. Time…….	**February 2011** Week 16 Date……. Time…….	**March 2011** Week 20 Date……. Time…….	**April 2011** Week 24 Date……. Time…….
Height (cm)					
BW (kg)					
BMI (kg/m^2)					
WC (cm)					
HC (cm)					
BMR (kcal)					
BF (%)					
FM (kg)					
FFM (kg)					
TBW (kg)					

APPENDIX 13

Researcher Record Sheet

Researcher record sheet

Name: **Age:**

Subject No. **Height**:

Variables / Date	Weight (kg)	BMI	% Body fat	Waist (cm)	Hip (cm)	Physical activity level
October 2010 Week1-Baseline						
January 2011 Week 12						
February 2011 Week 16						
March 2011 Week 20						
April 2011 Week 24						

APPENDIX 14

Consultation Record Sheet / Advice and Notes

Consultation record sheet / Advice and notes

Participant Name:

Subject No.

Date	Advice / Notes

APPENDIX 15

Participant record sheet /Goals and targets

Participant record sheet /Goals and targets

Name: **Subject No.**

Height:

Variables / Date	Weight (kg)	BMI (kg/m^2)	Body fat (%)	Waist (cm)	Hip (cm)	Physical activity level	Goals & Targets
October 2010 Week1-Baseline							
January 2011 Week 12							
February 2011 Week 16							
March 2011 Week 20							
April 2011 Week 24							

APPENDIX 16

5 A DAY Poster Guide

Just Eat More

Add flavour to a sandwich – throw in some lettuce and sliced tomato.

What counts?

- Fresh, frozen, chilled, canned, 100% juice, and dried fruit and vegetables all count.
- A portion of your 5 A DAY weighs approximately 80 grams, which is roughly a handful.
- Potatoes and other related vegetables such as yams and cassava do not count, because they are classified as starchy foods.
- The fruit and vegetables contained in convenience foods – such as ready meals, pasta sauces, soups and puddings – can contribute to 5 A DAY.
- Convenience foods can also be high in added salt, sugar or fat – which should only be eaten in moderation – so it's important to always check the nutrition information on food labels.

For more 5 A DAY information and tips, visit: nhs.uk/5aday

Eating 5 portions of fruit and vegetables every day may help reduce the risk of heart disease, stroke and some cancers.

nhs.uk/5aday

NHS

Just Eat More

For a healthier dessert try tinned peaches in their own juice.

Remember, frozen, canned, 100% juice, plus dried fruit and veg all count as well as fresh produce.

5 A DAY: what's it all about?

- **Eating a variety of fruit and vegetables, whether fresh, frozen, canned or dried, can all count towards your 5 A DAY. And, eating 5 A DAY may help to reduce the risk of heart disease, stroke and some cancers.**
- **Eating a variety of fruit and vegetables will give you plenty of vitamins and minerals. They are also a good source of fibre and other essential nutrients, all of which are important for your health.**

Just Eat More

For a healthier snack try dipping veg sticks into a dip.

Just Eat More

Have a glass (150ml) of 100% fresh juice with your lunch.

Are you getting your 5 A DAY?

1. How many portions of fruit* do you eat on a typical day? ☐ **Portions of fruit**
2. How many portions of vegetables* do you eat on a typical day? ☐ **Portions of vegetables**

(One portion = approximately 80 grams)

* See overleaf for examples of fruit and veg portion sizes.

Add up the numbers from your answers to questions 1 and 2: ☐

If the total is 5 portions or more, that's great. Remember, you need to eat a variety of fruit and vegetables.

If your total is less than 5 portions, then have a look at the 5 A DAY website for more hints and tips on how you can reach your 5 A DAY: **nhs.uk/5aday**

Just Eat More

Frozen fruit and veg count towards your 5 A DAY.

5 A DAY

Just Eat More
(fruit & veg)

nhs.uk/5aday

Look out for the 5 A DAY portion indicator on food packets

Where you see the portion indicator, it will feature how many portions of fruit or veg are in each serving.

5 A DAY

1 portion

5 A DAY

2 portions

Just Eat More

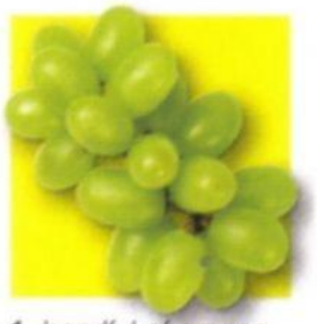
Feel like a snack? Reach for an apple instead of chocolate.

Eat a variety of fruit and vegetables, and aim for at least 5 A DAY.

1 medium apple

1 cereal bowl of mixed salad

2 halves of canned peaches



1 handful of grapes

1 medium banana

3 heaped tablespoons of peas

1 medium glass of orange juice

7 strawberries

3 whole dried apricots

5 A DAY

Just Eat More
(fruit & veg)

3 heaped tablespoons of cooked kidney beans

16 okra

NHS

APPENDIX 17

“Want to Lose Weight and Keep it off”

Want to Lose Weight and keep it off...?

First of all...

Write down your reasons for wanting to lose weight decide if this is the best time to make changes to your eating and activity - do you feel ready?

If now is the right time, before you start it really helps to:

- Learn more about your eating and activity habits. For about a week keep a record of everything you eat and drink as well as any activity you do. This helps you to identify problem areas, or feelings or situations that trigger overeating, and what you most want to change.
- Choose 2 or 3 small changes you can start with. Write yourself an action plan to follow with clear targets. For example; I will be more active and do this by doing a 10 minute walk three times each week. When successful you can move on to more changes or build on the ones you have already established.
- Make a list of things that will distract you from eating to use when you feel a bout of the munchies coming along.
- Enlist support. It not only keeps you inspired but helps you through tough times. It could be a friend, partner, health professional, group or website.

Set yourself realistic goals:

- Realistic goals are achievable, and success boosts confidence in your ability to lose weight.

- Losing just 5-10% of your weight over a 3-6 month period has massive health benefits.
- A weight loss of between 0.5-2lb a week is a safe and achievable target.
- It's not just about your weight on the scales. Losing inches from your waist really helps to lower the risk of conditions like type 2 diabetes and high blood pressure.
- Don't forget activity. Doing more every day not only helps you burn more calories but can boost mood and improve long term success.

Did you know? One pound of fat contains 3,500 calories, so to lose 1lb a week you need a deficit of 500 calories a day.

Follow a healthy eating plan:

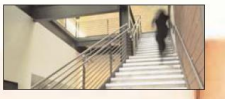
- Plan ahead to help ensure you have the right foods to hand, at the right times.
- Start the day with a healthy breakfast. People who do find it easier to control their weight.
- Eat regular, balanced meals. Try to have meals and snacks at planned times during the day.
- Aim to include at least five portions of fruit and vegetables each day - have some at every meal.
- Half fill your plate with veg/salad and divide the other half between meat, fish, egg or beans and carbs like potatoes, rice, pasta or bread.
- Choose foods and drinks that are low in fat and sugar and limit sweet and salty snacks.
- If you drink, moderate alcohol intake. It's high in calories and dissolves good intentions.
- Watch your portion sizes, especially when eating out.
- Avoid eating at the same time as doing something else, for example, when working, reading or watching TV.
- Eat slowly, concentrate on and really taste what you are putting in your mouth.
- Finally, it takes time for your brain to know your stomach is full so wait at least 5 -10 minutes before deciding if you need more.

Be more active:

Moving your body around means using up more calories than if you are sitting down - every little helps...

- Look for easy ways to fit more movement into your day-to-day routine. For example, stand up when on the phone, use a toilet further away, park a bit further away from your destination or get off the bus a stop earlier.
- Plan a walk into your day, perhaps ask someone in your

family or a friend to walk with you. Gradually increase the length of time and the speed of your walks to burn more calories.



- Try using a step counter or pedometer to see what you currently manage then gradually add to the number of steps you do - build up to 10,000 a day - or more.

- Slowly build on the amount of activity that you do so that it becomes part of your daily routine, not just a passing phase that you find too difficult to keep up. People who do are far more successful with long term weight control.

- Plan your activities into your diary each week. Tick it off and be proud when you have done it.

Follow an eating and activity plan that is tailored to you as an individual and to your lifestyle - you are much more likely to stick to it.

While you are on your weight loss journey:

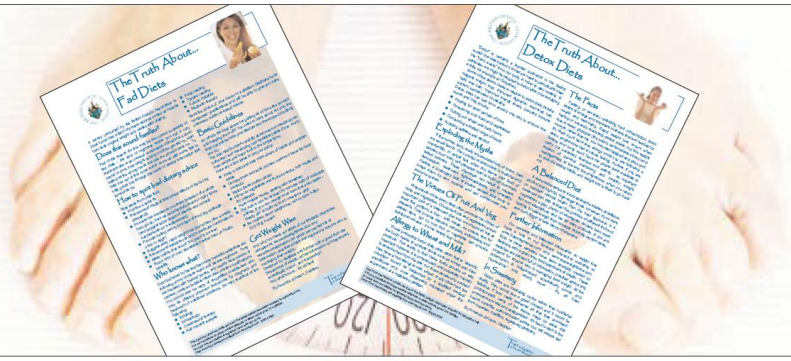
- Accept that the occasional lapse is normal and get straight back on track. Don't let it make you lose sight of your overall goals and the progress you have made. Instead learn from it to help you in the future.
- Keep some form of food diary. People who do, lose more weight. It not only helps you to stay aware of what you are doing, and deal with any problems like comfort eating, but helps you feel in control of your eating.
- Track your progress; you might want to do this by weighing yourself (at least 1-2 times a week), measuring your waist, reviewing your food diary or seeing what goals you have achieved at the end of each week.
- Plan for events like eating out and parties. Decide what your plan of action will be and stick to it so that you can enjoy your night out For example, when eating out decide to have two courses instead of three or share a pudding that you would normally eat to yourself.

- Making changes to your lifestyle is hard work. When you achieve a goal reward your success with something you value (e.g. magazine, CD, trip to the cinema).

- Keep in touch with your supporters - it makes all the difference. Decide who can help you and tell them the best way they can do it (e.g. ask a friend to go to an exercise class with you, ask your partner to not buy you food as a gift).

Remember there is no quick fix. People who successfully lose weight and keep it off stay realistic and develop the skills to make their new eating and activity habits an enjoyable way of life.

Check out other Fact Sheets for information on Fad Diets and Detox Diets:



Why not have a look at our websites www.bdaweightwise.com and www.teenweightwise.com which are packed full of trustworthy information, further advice and support for anyone wanting to lose weight.