IRON DEFICIENCY AND OBESITY

Are the two linked?

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Abstract

Introduction and background: The inverse relationship between iron status and obesity, contributes to the global double burden of malnutrition. The aim of this research was to investigate the potential links between obesity and iron deficiency (ID), particularly in women of child-bearing age.

Objectives: To assess the effectiveness of interventions designed to address ID in women who are either overweight or obese, to explore the effectiveness of interventions designed to reduce inflammation, to identify any links between BMI, iron status, fatigue level and diet and to increase depth of information available regarding the fatigue experience.

Methods: A mixed methodological approach was undertaken with two systematic style reviews, two quantitative studies and one qualitative study. The reviews were designed to fulfil the first two objectives with data reported using the PRISMA framework for transparency. In the first quantitative study new Slimming World (SW) female members were invited to take part in an online survey including validated tools to determine levels of iron intake and fatigue levels. The second quantitative study was modelled on the first study, but with cultural/practical adjustments and the availability of iron-related blood parameters. Turkish women, who consulted a registered dietitian to lose weight, were invited to take part in a paper-based survey. A qualitative study compromised of two methodological phases with a focus group and telephone interviews conducted.

Results: The first review found that although intervention trials which assess iron status are available, to date, no study has been undertaken where the primary aim is to improve iron status of women who are obese. Effective interventions included additional dietary iron in a very low energy diet, higher protein and energy restriction, breakfast cereals as part of a weight loss intervention and exercise training. The second review found that weight loss may be associated with a decline in some biomarkers of inflammation, such CRP, in adult women who are obese. Also, a higher protein, brown rice, aerobic exercise, histidine supplementation, and tomato juice were found to be effective dietary strategies in reducing inflammation with or without weight loss.

The first quantitative study found that SW is an effective weight management programme in helping to achieve reductions in BMI, fatigue levels and consumption of energy-dense foods, and increasing PA levels and consumption of high-fibre/antioxidant foods. The second quantitative study further supported Study 1 and additionally the relationship between iron status and low-grade inflammation in women who are obese was reported. The qualitative study suggests most women in the study believed that fatigue had negative effects on their perceived ability to make lifestyle changes.

Conclusion: Weight management has a positive impact on reducing inflammation, improving iron status, fatigue level, quality of life and motivation/ability to make lifestyle changes in women of reproductive age who are obese.

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Dedication

To my husband Ömer Furkan and my son Bilge Kağan

This humble work is a sign of my love to you!

List of publications & communications

Oral communications

1. Kaya Kacar, H., Bennet, S., Avery, A. & McCullough, F. Does engagement in a weight management programme improve level of fatigue? International Conference on Obesity and Diabetes, July 10-12, 2017, Dubai, UAE (Appendix A)
2. Kaya Kacar, H., Bennet, S., Avery, A. & McCullough, F. Associations between body weight, fatigue and levels of physical activity in non-pregnant women accessing a weight management programme. Women and Maternal Nutrition and Care, August 31-September 01, 2018, Toronto, Canada (Appendix B)

Posters at conference

1. Kaya Kacar, H., Avery, A. & McCullough, F. Iron deficiency and obesity. 17th International Congress of Dietetics, September 7-10, 2016, Granada, Spain (Appendix C and D)
2. Kaya Kacar, H., Bennet, S., Avery, A. & McCullough, F. Iron deficiency anaemia (IDA) and obesity – Are the two conditions linked? ASO UK Congress on Obesity 2016, September 19-20, Nottingham, UK (Appendix E and F)
3. Kaya Kacar, H., Bennet, S., Avery, A. & McCullough, F. Relationship between fatigue and level of physical activity in women who engaged in

a weight management programme. ASO UK Congress on Obesity 2017, September 7-8, Pontypridd Cardiff, UK (Appendix G and H)

4. Kaya Kacar, H., Bennet, S., Avery, A. & McCullough, F. Impact of a weight management programme on the dietary habits of non-pregnant women. Nutrition Society Summer Conference 2018: Getting energy balance right, July 10-12, Leeds, UK (Appendix I and J)
5. Kaya Kacar, H., Avery, A. & McCullough, F. A hidden danger of obesity: Iron deficiency anaemia. Women and Maternal Nutrition and Care, August 31-September 01, 2018, Toronto, Canada (Appendix K)

Journal articles submitted

1. Strathearn, L., Kaya Kacar, H. & Avery, A. Changes in dietary patterns with engagement in a weight management programme: can SACN's recommendations be met?

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List of abbreviations and acronyms

Apo-Tf	Apo-transferrin
BMI	Body mass index
BMPR	Bone morphogenetic protein receptor
BSW	Before joining Slimming World
CD	Cluster of differentiation
CHO	Carbohydrate
CI	Confidence interval
Cp	ceruloplasmin
CRP	C-reactive protein
DcytB	Duodenal cytochrome b
Dmt1	Proton-coupled divalent metal transporter 1
EAR	Estimated average requirement
ERK	Extracellular signal-regulated kinases
Fe^{2+}	Ferrous
Fe^{3+}	Ferric
FeFFQ	Iron-food frequency questionnaire
FFQ	Food frequency questionnaire

Fpn1	Ferroportin-1
GDF-15	Growth differentiation factor 15
GFI	Global fatigue index
Gpi	Glycosylphosphatidylinositol
HAMP	Hepcidin gene
Hb	Haemoglobin
HCP1	Haem-carrier protein 1
Heph	Hephaestin
Hjv	Hemojuvelin
HO1	Hemeoxygenase1
hs-CRP	High-sensitivity C-reactive protein
ID	Iron deficiency
IDA	Iron deficiency anaemia
IL	Interleukin
IL-6	Interleukin-6
IPAQ	International physical activity questionnaire
IREG1	Iron regulated transporter
KMO	Kaiser-Meyer-Olkin Measure of Sampling Adequacy

LRNI	Lower reference nutrient intake
MAF	Multidimensional Assessment of Fatigue
MAPK	Mitogen-activated protein kinases
MET	Metabolic Equivalent of Task
mRNA	Messenger ribonucleic acid
MT2	Matriptase-2
MTF-1	Metal-responsive transcription factor-1
OR	Odds ratio
PA	Physical activity
PCA	Principal component analysis
PCBP1	Poly binding protein 1
RBCC	Red blood cell concentrate
RDA	Recommended Dietary Allowance
RNI	Reference nutrient intake
sCp	Serum ceruloplasmin
SD	Standard deviation
SE	Standard error
SLC40A1	Solute carrier family 40 member 1

SPSS	Statistical Package for the Social Sciences
sTfR	Soluble transferrin receptor
SW	Slimming World
Tf	Transferrin
TfR	Transferrin receptor
TIBC	Total iron binding capacity
TNF-α	Tumour necrosis factor-α
TWSG-1	Twisted gastrulation 1
VLED	Very low energy diet
WHO	World Health Organisation

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CHAPTER 1: SETTING THE SCENE

This PhD explores the relationship between "iron deficiency and obesity". In this chapter, the prevalence of these issues globally, as well as in both the UK and Turkey is summarised. The relationship between iron status and obesity and the role of inflammation and fatigue in obesity have been of increasing interest to clinicians over the past several decades. Knowledge gaps in this field are identified and the aims of the studies and the research questions are stated. The chapter ends with an overview of the research study chapters that comprise this thesis.

1.1 LITERATURE REVIEW

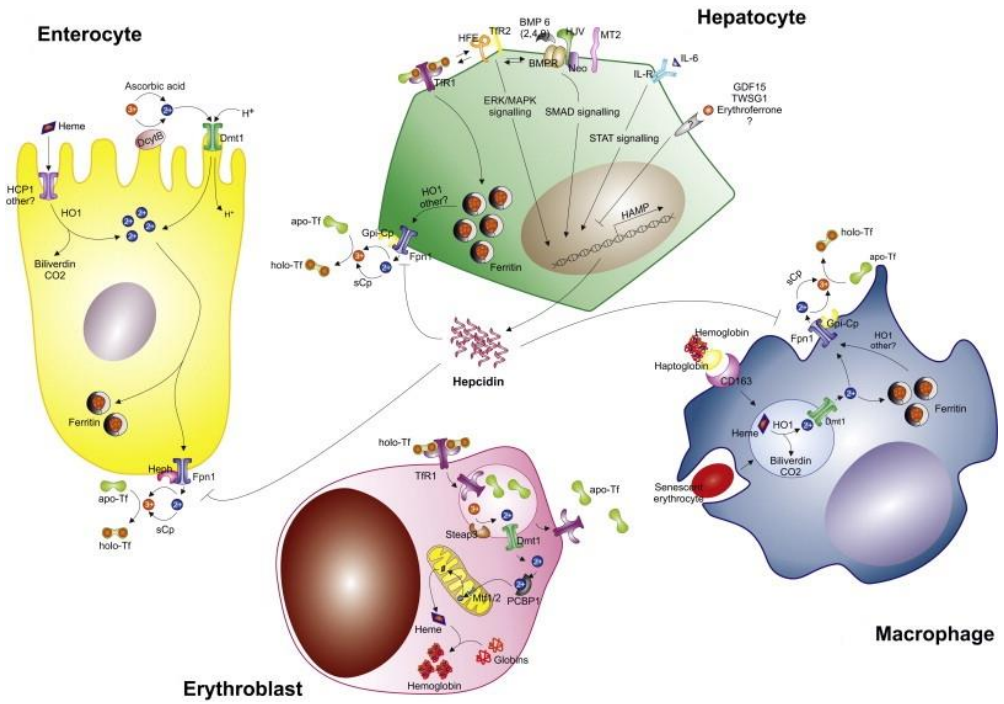
1.1.1 Iron metabolism in the human body

Iron is an essential micronutrient for a number of vital biological processes including oxygen transport, electron transfer and catalytic reactions (Papanikolaou & Pantopoulos, 2005). Ferrous (Fe^{2+}) and ferric (Fe^{3+}) are the most common forms of iron in the human body. To assist in the different metabolic functions, and because iron has toxic effects when found freely, it is attached to a number of different proteins. In serum it is coupled with transferrin; in the cell, chaperons deliver iron to ferritin and it is stored as ferritin (Silva & Faustino, 2015). Some peptides, including iron regulatory proteins, hepcidin and matriptase, play a role in iron regulation at different physiological levels. Other proteins, including myoglobin and haemoglobin (Hb), are the final products of iron metabolism as iron is required for their function (Waldvogel-Abramowski *et al*., 2014; Weiss, 2009).

1.1.1.1 Regulation of iron homeostasis

Maintenance of iron homeostasis includes a number of processes. The majority of the oxidised form of iron (Fe^{3+}) is reduced to Fe^{2+} by ascorbic acid and duodenal cytochrome b at the apical membrane of enterocytes. Reduced iron is absorbed by proton-coupled divalent metal transporter 1 and driven to the basolateral membrane of enterocytes. After being driven, ferroportin-1 exports iron for circulation in association with transferrin. Transferrin is endocytosed by erythrocytes. After being endocytosed, mitochondria use iron in order to synthesise haem. Haem is incorporated in haemoglobin which is responsible for transporting oxygen. When excessive iron is absorbed it is stored in enterocytes as ferritin in the hepatocytes. Iron is recycled by macrophages. Old erythrocytes are ingested and destroyed and then the macrophages release iron and make it available for re-utilisation. All of these processes are controlled by hepcidin (see figure 1.1) (Silva & Faustino, 2015).

Figure 1.1 Systemic iron metabolism regulation (Silva & Faustino, 2015)



Apo-Tf, apo-transferrin; **BMPR**, bone morphogenetic protein receptor; **CD**, cluster of differentiation; **Cp**, ceruloplasmin; **DcytB**, duodenal cytochrome b; **Dmt1**, proton-coupled divalent metal transporter 1; **ERK**, extracellular signal-regulated kinases; **Fpn1**, ferroportin-1; **GDF-15**, growth differentiation factor 15; **Gpi**, glycosylphosphatidylinositol; **HAMP**, hepcidin gene; **HCP1**, haem-carrier protein 1; **Heph**, hephaestin; **Hjv**, hemojuvelin; **HO1**,hemeoxygenase1; **IL**, interleukin; **MAPK**, mitogen-activated protein kinases; **MTF-1**, metal-responsive transcription factor-1; **MT2**, matriptase-2; **PCBP1**, poly binding protein 1; **sCp**, serum ceruloplasmin; **Tf**, transferrin; **TfR**, transferrin receptor; **TWSG-1**, twisted gastrulation 1.

1.1.1.1.1. Hepcidin

Hepcidin was originally known as liver-expressed antimicrobial protein 1. It is a 25-amino acid peptide hormone considered as a main regulator of body iron metabolism (Schmidt, 2015). Although hepcidin is produced mainly by the liver, macrophages and cells from non-hepatic tissues including heart, brain and adipose tissue express it in small amounts (Papanikolaou & Pantopoulos, 2017).

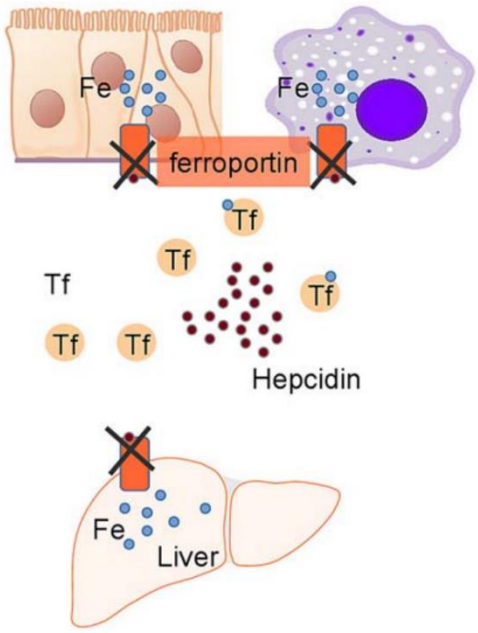
There are three primary sources of iron that are controlled by hepcidin: dietary absorption in the duodenum, recycled iron released from macrophages and stored iron released from hepatocytes (Ganz & Nemeth, 2012). A number of stimulus factors regulate plasma hepcidin levels. The main regulators of hepcidin secretion by the liver are plasma and liver iron levels, erythropoiesis and inflammation (Girelli *et al*., 2016). Hepcidin is increased in response to high plasma and liver iron to maintain stable body iron levels. Low iron and erythroid activity inhibit hepcidin expression to provide iron supply for erythropoiesis (see figure 1.2) (Girelli *et al*., 2016).

Concentrations of hepcidin and certain defensins (cysteine-rich cationic proteins which play an active role against bacteria, fungi, and some types of viruses) increase similarly during infection and inflammation to restrict the use of extracellular iron by microbes (see figure 1.3) (Barrera et al., 2012; Ganz & Nemeth, 2015). In addition, there is a direct anti-microbial activity of hepcidin *in vitro*, like defensins. Due to these strong connections, hepcidin is considered as a mediator of host defence. More details about the effect of inflammation on hepcidin expression is discussed in separate paragraphs: “Inhibiting factors of

iron absorption" and "Obesity-related inflammation and reduced iron absorption".

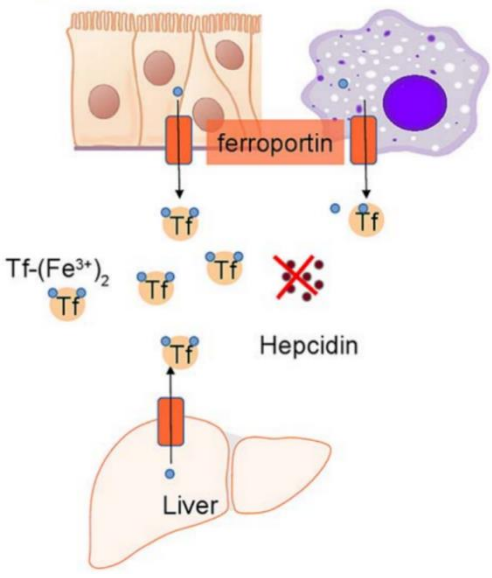
In respect to systemic iron homeostasis, hepcidin restricts the iron flow into the plasma by binding to ferroportin (Papanikolaou & Pantopoulos, 2017). Ferroportin is also called solute carrier family 40 member 1 (SLC40A1) or iron regulated transporter (IREG1). It is the cellular exporter of iron and bound by hepcidin which induces its endocytosis and degradation. Ferroportin is found in not only macrophages and hepatocytes but also all the tissues where major iron flows are regulated. Ferroportin has no alternative and it is the only cellular way out for iron (Ganz, 2005)

Figure 1.2 Hepcidin-mediated regulation of iron efflux to the bloodstream in response to high iron or inflammatory signals (Papanikolaou & Pantopoulos, 2017).



Tf, transferrin.

Figure 1.3 Hepcidin-mediated regulation of iron efflux to the bloodstream in response to low iron or high erythropoietic drive (Papanikolaou & Pantopoulos, 2017).



Bioavailability of iron refers to the percentage of systemically used iron that is absorbed and transferred into the organism by the intestinal mucosa (Geissler & Singh, 2011). Iron bioavailability is influenced by the chemical form of iron, either haem or non-haem. Haem iron is highly abundant in animal foods attached to haemoproteins, haemoglobin and myoglobin (Silva & Faustino, 2015). Non-haem iron is found in plant-based foods such as certain fruits, vegetables, legumes, grains and nuts (Nair & Augustine, 2018).

The majority of dietary non-haem iron is in the Fe^{3+} form. Because this form of iron is insoluble, it first needs to be reduced to the Fe^{2+} form (Kapil, 2017). The membrane bound ferric reductase duodenal cytochrome B is expressed on the apical brush border membrane of intestinal epithelial cells to convert Fe^{3+} to Fe^{2+}. Then, the divalent metal transporter carries Fe^{2+} across the apical membrane of enterocytes. Iron enters the systemic circulation by crossing the basolateral membrane of intestinal enterocytes by ferroportin (Wallace, 2016).

Prentice (2017) has recently summarised knowledge about iron absorption:

I. There is an active auto-regulatory feedback mechanism in the absorption of iron whereby absorption can easily be increased when there is a greater demand e.g. in pregnancy.

II. Absorption and utilisation of haem iron is better than non-haem iron.

III. There are some binding agents in foods including phytates and phenolic compounds that inhibit iron absorption.

IV. Co-ingestion of vitamin C enhances iron absorption.

V. Iron and zinc compete for absorption and thus negatively affect the absorption of each other.

VI. A physiological regularity mechanism is not available for the excretion iron from the body.

1.1.2 Enhancing factors of iron absorption

1.1.2.1 Dietary factors

The bioavailability of haem iron is relatively high (15%-35%) and its absorption is influenced by other dietary factors to only a small extent. In contrast the bioavailability of non-haem iron is much lower (2%-20%) and its absorption is strongly affected by the presence of some food components (Hurrell & Egli, 2010).

Several dietary factors including the type of foods, type of diet and methods of food preparation can affect iron absorption positively (Kapil, 2017).

Ascorbic acid, also known as vitamin C, is one of the significant enhancers of the bioavailability of iron and its enhancing effect on iron absorption has been explored in several studies (e.g., Hou *et al*., 2016; Singh & Prasad, 2018). In terms of the underlying mechanism, it is commonly reported that ascorbic acid promotes iron absorption in the duodenum by inducing reduction of dietary ferric iron to more soluble and absorbable ferrous form (Gulec *et al*., 2014). Additionally, Lane & Richardson (2014) suggest that cellular iron uptake and metabolism in the mammalian system can be regulated by ascorbic acid through the stimulation of ferritin synthesis which prevents lysosomal ferritin degradation and reduces cellular iron efflux. Also, the transferrin-iron uptake

that supplies almost all cellular iron under physiological conditions is stimulated by ascorbic acid (Lane *et al*., 2013).

Meat, poultry, fish and other sea foods are also enhancers of non-haem iron absorption (Kapil, 2017). Reddy *et al*., (2006) investigated the effect of meat consumption on non-haem iron absorption. In the study, iron absorption was measured by having the healthy subjects (n=14) ingest a radioiron-labelled bread. Although there was an increase in non-haem iron absorption from 4.8% to 6.5% with a high-meat diet, this difference was not statistically significant ($p=0.075$) which may be partially explained by the small study population.. However further work supports the observation that haem iron absorption is not influenced by the presence of animal proteins. Pizarro *et al.* (2016) measured the effects of dietary animal proteins on the absorption of haem iron. Healthy women aged 35-45 years (n=45) received the same dose of 5 mg of iron in gelatine capsules. The capsules contained different sources of iron including haem, red blood cell concentrate (RBCC), Hb, RBCC + beef meat, haem + fish, chicken, beef and purified animal protein (casein, collagen, albumin). It was showed that the bioavailability of haem iron significantly increased when ingested as RBCC (nearly 2-fold) or RBCC + beef (1.6-fold) ($p<0.05$). However, its absorption significantly reduced when it was blended with fish and chicken ($p<0.05$) and did not change when presented with casein, collagen, and albumin. It was concluded that haem iron absorption was not enhanced by the proteins from different animal source foods and their digestion products. Similarly, Weinborn *et al*., (2015) determined the effect of proteins from cereals (zein, gliadin and glutelin) and legumes (soy, pea and lentil) on haem iron absorption. Healthy women aged 35-45 years (n=30) ingested the same dose of

5 mg of iron in gelatine capsules containing haem only and haem plus the proteins. Iron absorption was determined by antecubital venous sample and it was found that haem iron absorption was not affected by the different cereal proteins. The legume proteins, including pea and lentil, had no effect on haem iron absorption but soy proteins decreased haem iron absorption ($p<0.05$).

In a recent study (Weinborn *et al*., 2017), the effect of a prebiotic mix on haem and non-haem iron bioavailability was examined. In this randomised controlled study, intervention participants consumed one yogurt with a prebiotic mix and control participants, one yogurt without the prebiotic mix per day for 12 days. Radioactive iron was incorporated into erythrocytes and its absorption was measured. The results suggested that haem and non-haem iron bioavailability did not change in the control group. However, there was a significant increase in haem iron bioavailability (by 56%, $p<0.007$) after the consumption of prebiotic. A significant difference was not found in non-haem iron bioavailability in the prebiotic group. It was concluded that although daily prebiotic mix intake has no effect on non-haem iron bioavailability, it increases bioavailability of haem iron. One might hypothesise that components such as acetic, propionic, and butyric acids, which are produced after prebiotic fermentation, reduce the pH of colon contents and enhance iron absorption (Coudray *et al*., 1997).

Other dietary factors that affect iron bioavailability are food preparation and processing methods. Phytic acid and hexa- and penta-inositol phosphate inhibit iron absorption but can be decreased by cooking, fermentation, or germination due to thermal and enzymatic action. Reduced phosphate groups can improve

bioavailability of non-haem iron (Kapil, 2017). In order to assess the effects of lactic fermentation on the bioavailability of iron from a vegetable mix, Scheers *et al*. (2016) conducted a study. In this study a mix of fresh or fermented vegetables consisting of carrots, turnips, cabbage, parsnip, celery, and onion was given to the participants with the low-phytate meal (white wheat rolls) and with the high-phytate meal (wheat bran rolls). The results showed that there was an increase in iron absorption from 13.6 ± 1.6 to 23.6 ± 2.0 % ($p < 0.0001$) and from 5.2 ± 0.8 to 10.4 ± 2.4 % ($p < 0.0001$) in the low and high phytate meal respectively when fermented vegetables replaced fresh vegetables. It was suggested that iron absorption is increased by lactic-fermented vegetables, possibly due to an increase in hydrated ferric iron (available ferric iron in the digested fermented samples)

1.1.2.2 Host factors

The absorption of dietary iron is tightly regulated according to body iron requirements and is influenced by the health and iron status of the person. More iron is absorbed by people who have iron deficiency (ID) (Kapil, 2017). Fuqua *et al*. (2012) reported that adequate dietary iron must be absorbed in case of obligate losses including deprivation of epithelial cells and menstruation to meet iron requirements. Also, because iron requirements increase during growth, hypoxic conditions, and after blood loss, iron supply needs to respond.

Pregnancy is another period that iron stores and intake are insufficient to overcome increased iron requirements (Moll & Davis, 2017). Since the developing foetus needs iron and maternal erythroid mass expands, iron demand and absorption is high (Gulec *et al*., 2014). Also, more iron is required by

postnatal woman and the neonate with iron absorbed from breast milk very efficiently (ibid).

1.1.3 Inhibiting factors of iron absorption

1.1.3.1 Dietary factors

Dietary components that inhibit iron absorption are phytates, phenolic components including tannins, and soy proteins (Kapil, 2017).

Phytates are present in bran, oats and fibre-rich foods as a storage form of phosphates and minerals (Zijp *et al*., 2000). Although phytates are considered as the main inhibitors of iron absorption (Hurrell & Egli, 2010), it was found that the negative effect of phytate on non-haem iron absorption can be decreased by habitual consumption of a high-phytate diet among women with suboptimal iron stores (Armah *et al*., 2015). In a parallel design study, women with suboptimal iron stores (serum ferritin ≤30 μg/l, n=32) were randomly assigned to high-phytate and low-phytate groups. At baseline, the serum iron response over 4 hours after a test meal containing 350 mg of phytate was measured. During the eight-week dietary intervention, participants consumed either high-phytate (mean phytate concentration: 590 mg/100 g) or low-phytate foods (mean phytate concentration: 90 mg/100 g) with at least 2 of their daily meals. After the intervention the same test meal was consumed by the participants and it was shown that the serum iron response to the test meal increased in the high-phytate group (41% increase in the area under the curve). However, no effect was observed in the low-phytate group (21% decrease in the area under the curve) (ibid).

Phenolic compounds bearing catechol or galloyl groups are released during digestion from food or drinks including tea, coffee, cocoa, red wine, many vegetables (spinach, aubergine and beetroot greens), grains (red sorghum), herbs and spices (oregano, cinnamon) (Zijp *et al*., 2000; Mascitelli & Goldstein, 2014). In the intestinal lumen, iron can combine with them and non-haem iron absorption can be strongly inhibited (Mascitelli & Goldstein, 2014). In terms of haem iron absorption, the effects of epigallocatechin gallate and grape seed extract on haem iron absorption was investigated and it was found that bioactive dietary polyphenols almost completely blocked haem iron absorption by reducing basolateral iron efflux in human intestinal Caco-2 cells (Ma *et al*., 2010).

Tea is a polyphenol-rich beverage and extensively consumed worldwide (Fang *et al*., 2003). In 2016, the UK was the third largest tea-consuming country in the world (4.28 pounds per capita per year) and Turkey was ranked first with an annual tea consumption of 6.96 pounds per capita (Statista, 2019). In Turkey, tea-drinking is considered a food-culture and Turkish people consume tea all day long starting from morning until bed time (Chambers *et al*., 2016).

In the past two decades a number of researchers have reported that tea is a potent inhibitor of non-haem iron absorption in both iron-deficient and non-iron deficient individuals (Hurrell *et al.,* 1999; Thankachan *et al*., 2008; Samman *et al*., 2011). These results are consistent with those of Ahmad Fuzi (2017) who reported the negative effects of tea consumption on non-haem iron absorption in non-anaemic women. However, it was found that iron absorption was significantly higher (2.2%) when tea was consumed one hour after the meal than

when tea was consumed simultaneously with the meal (p=0.046). It was concluded that the inhibitory effect of tea can be reduced by a 1-h time interval between a meal and tea consumption.

1.1.3.2 Host factors

Infections and inflammation influence iron absorption negatively by increased hepcidin expression (Hurrell & Egli, 2010). During infections, hepcidin (mediator of host defence) increases within hours and concentration of plasma iron reduces acutely. It is likely that the supply of iron to microbial invaders is restricted by lowered plasma and extracellular iron concentration (Ganz, 2015). This is an important physiological response of human antimicrobial defence because iron is an essential nutrient for not only host but also pathogens (Cherayil, 2011). The flow of iron to the bone marrow and erythropoiesis are limited by increased circulating hepcidin levels and this results in the characteristic anaemia associated with infection and inflammation (Ganz, 2015). This type of anaemia is known as anaemia of inflammation or anaemia of chronic disease whose defining biochemical features include low serum iron despite adequate systemic iron stores (Nemeth & Ganz, 2014).

Obesity is an inflammatory disorder and associated with decreased iron absorption (Cepeda-Lopez *et al*., 2010). The effect of overweight and obesity on iron absorption and metabolism is discussed in a separate section.

1.1.4 Dietary recommendations for iron

The UK recommended intake for iron and other nutrients were revised by the Committee on Medical Aspects of Food and Nutrition (COMA) in 1991

(Department of Health, 1991) and reviewed by the Scientific Advisory Committee on Nutrition (SACN) in 2010. In the COMA report, Dietary Reference Values (DRVs) were presented as Lower Reference Nutrient Intake (LRNI), Estimated Average Requirement (EAR) and Reference Nutrient Intake (RNI) to provide benchmark levels of nutrient requirements. The EAR refers to the mean requirement for the population and the RNI is defined as two notional standard deviations above the EAR. The LRNI is defined as two notional standard deviations below the EAR and represents the amount of a nutrient that meets the requirements of just 2.5% of the population. The RNI for iron is 8.7 mg/day for men and women aged over 50, and 14.8 mg/day for premenopausal women (aged between 11-50 years) (Scientific Advisory Committee on Nutrition, 2010) (see table 1.1).

Table 1.1 Dietary reference values for iron presented as mg/day (µmol/day) (Department of Health, 1991)

Age	LRNI	EAR	RNI
15-18 years (M)	6.1 (110)	8.7 (160)	11.3 (200)
15-18 years (F)	8.0 (140)	11.4 (200)	14.8 (260)
19-50 years (M)	4.7 (80)	6.7 (120)	8.7 (160)
19-50 years (F)	8.0 (140)	11.4 (200)	14.8 (260)
50+ years	4.7 (80)	6.7 (120)	8.7 (160)

M, males; **F**, females

1.1.5 Iron deficiency and iron deficiency anaemia

The World Health Organisation (WHO) (2001) defined ID as "a condition in which there are no mobilizable iron stores and in which signs of a compromised supply of iron to tissues, including the erythron, are noted". WHO (2014) defined anaemia as "a condition in which the number and size of red blood cells, or the haemoglobin concentration, falls below an established cut-off value, consequently impairing the capacity of the blood to transport oxygen around the body". Anaemia in non-pregnant women (above 15 years of age) is defined as Hb <120 g/l (WHO, 2001) (see table 1.2).

Table 1.2 Haemoglobin and haematocrit levels below which anaemia is present in adult population (WHO, 2001)

Age or gender group	Haemoglobin	Haematocrit	
	g/l	mmol/l	l/l
Non-pregnant women (above 15 years of age)	120	7.45	0.36
Pregnant women	110	6.83	0.33
Men (above 15 years of age)	130	8.07	0.39

There are two main forms of ID: absolute or functional. Absolute ID occurs in the case of low or exhausted body iron stores. If total body iron stores are normal or elevated despite inadequate iron supply to the bone marrow, functional ID arises. These two types of ID can coexist (Lopez *et al*., 2016).

ID can exist with or without anaemia. However, even mild and moderate forms of iron deficiency anaemia (IDA) may affect a number of factors including

cognitive development, immunity, learning ability and work capacity (Beard & Connor 2003; Abbaspour *et al*., 2014). Whilst some symptoms of IDA are related to all anaemias and these 'subjective' symptoms can include pallor of the skin, conjunctivae, and nail beds, other symptoms are more objective and relate to the impaired hypoxic functioning. These objective symptoms include fatigue, exertional dyspnoea progressing to breathlessness at rest, vertigo, syncope, headache, tachycardia, and a cardiac systolic flow murmur (Lopez *et al*., 2016). Lopez *et al.* (2016) revised the symptoms of IDA according to the frequency of the cases (see table 1.3).

Table 1.3 Symptoms of IDA (Lopez et al., 2016 and references therein)

Very frequent	Frequent	Rare
Headache (63%)	Diffuse and moderate alopecia (30%)	Haemodynamic instability (2%)
Fatigue (44%)	Atrophic glossitis (27%)	Syncope (0.3%)
Dyspnoea[+]	Restless legs syndrome (24%)	Koilonychia[+]
Paleness (45-50%)	Dry and rough skin[+]	Plummer-Vinson syndrome (<0.1%)
	Dry and damaged hair[+]	
	Cardiac murmur (10%)	
	Tachycardia (9%)	
	Neurocognitive dysfunction[+]	
	Angina pectoris[+]	
	Vertigo[+]	

[+] Percentages of the frequency for these symptoms were not available in the reference (Lopez et al., 2016).

The frequency of fatigue as a burdensome symptom in IDA is clearly supported by a qualitative study which evaluated the content validity of the Functional Assessment of Chronic Illness Therapy - fatigue scale in IDA patients (Acaster *et al*., 2015). Qualitative interviews included 15 patients and it was indicated that all patients spontaneously reported “fatigue”, “tiredness”, and/or “low energy” as their main symptom experiences and primary symptoms that influenced their health-related quality of life.

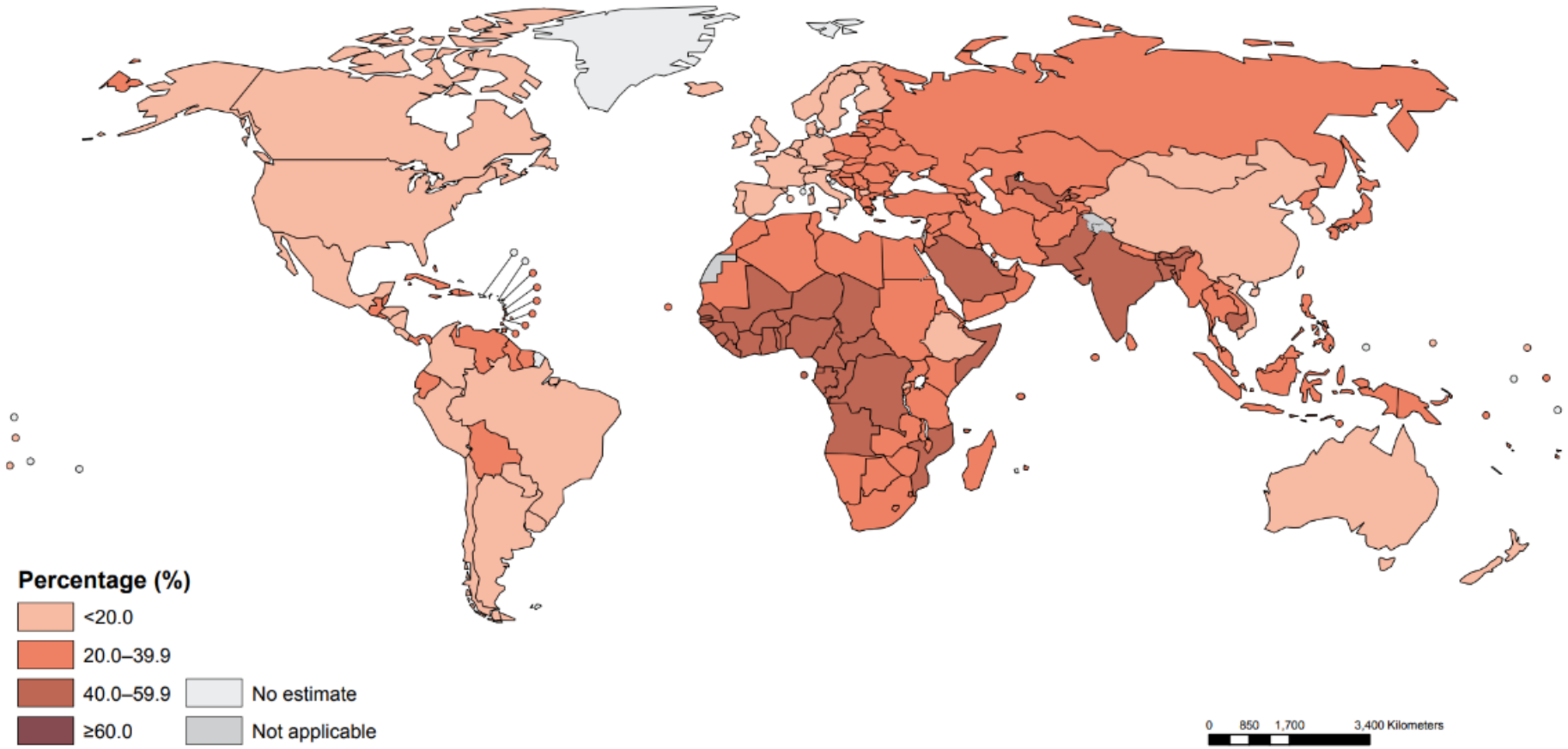
Fatigue, one of the most common symptoms of IDA, was also found associated with ID without anaemia. A cross-sectional study (Sawada *et al*., 2014) assessed the relationship between ID without anaemia and mental and somatic symptoms including anger and fatigue in young Japanese women of reproductive age (18-22). ID without anaemia was defined as having Hb ≥ 12 g/dl and serum ferritin < 20 ng/ml. Anger (p=0.007) and fatigue (p=0.017) were significantly higher in women who had ID but not anaemia than those who had normal iron status. Patterson *et al*. (2001) found that fatigue can be decreased by treatment of ID with either supplementation or an iron-rich diet in women of childbearing age who have iron deficiency. Similarly, Krayenbuehl *et al*. (2011) reported that fatigue can be improved by intravenous administration of iron in non-anaemic women with low serum ferritin concentration.

1.1.5.1 The global prevalence of iron deficiency anaemia

In a systemic review by Petry *et al*. (2016), the prevalence of ID, IDA, and anaemia among pre-school children and non-pregnant women of reproductive age was reported. Data from 23 countries ranked as low, medium, and high by the Human Development Index was analysed. Among non-pregnant women of

reproductive age, the overall mean prevalence of ID, IDA and anaemia was 20.8%, 10.8% and 26.6%, respectively. Whilst the highest prevalence of IDA among non-pregnant women was reported in Central Asia (13.7%), the lowest prevalence was reported in South, East, and Southeast Asia (7.8%). WHO (2015) illustrated the classification of countries by degree of public health significance of anaemia, based on blood Hb concentration in 2011 for non-pregnant women of reproductive age by map (see figure 1.4).

Figure 1.4 Prevalence of anaemia, non-pregnant women aged 15-49 years, 2011 (WHO, 2015)



According to WHO (2015), the mean Hb concentration of non-pregnant women aged 15-49 years in the UK was 130 g/l. Among the same group, the percentage of Hb less than 120 g/l and 80g/l was 14% and 0.2%, respectively.

In 2012, member states of the WHO endorsed six global nutrition targets for 2025 in order to improve maternal, infant and young-child nutrition. The second global nutrition target for 2025 is "a 50% reduction of anaemia in women of childbearing age (15-49 years)" (WHO, 2012). According to the WHO (2014), this reduction could be considered equal to a 5.3% reduction per year between 2012 and 2025 and this corresponds with approximately 230 million anaemic non-pregnant women.

1.1.5.2 The prevalence of iron deficiency anaemia in Turkey

As Turkey does not have national or subnational data on the prevalence of anaemia, WHO (2015) reported estimated mean blood Hb concentration and the percentage of anaemia for Turkey. According to this report, the estimated mean Hb concentration (g/l) was 127, 118, and 126 for non-pregnant women (15-49 years), pregnant women (15-49 years) and all women of reproductive age (15-49 years), respectively.

The estimated percentage of non-pregnant women with blood Hb concentration <120 g/l and <80 g/l were reported as 29% and 1.0%. The estimated percentage of pregnant women with blood Hb concentration <120 g/l and <80 g/l were reported as 28% and 0.3% (WHO, 2015). However, these findings were inconsistent with a descriptive study (Saydam *et al*., 2017) that determined the prevalence of anaemia and related factors among women (15-49 years) in Turkey. Saydam *et al*. (2017) observed that the percentage of ID, IDA (Hb<120

g/l) and severe anaemia (Hb<70 g/l) were 15.6%, 10.3% and 1.9% respectively among Turkish women.

1.1.6 Overweight and obesity

The WHO (2018) defined overweight and obesity as abnormal or excessive body fat that may negatively affect health. To classify overweight and obesity in adults, the body mass index (BMI) is used as a pragmatic measure. In order to calculate BMI, a person's weight (kg) is divided by the square of their height (m) (WHO, 2018). A BMI ≥ 25 kg/m^2 is defined as overweight and a BMI ≥ 30 kg/m^2 is defined as obesity by WHO (2018).

An energy imbalance between intake and expenditure is the main cause of overweight and obesity. The consumption of energy-dense foods and physical inactivity has been increasing globally (WHO, 2018).

Overweight and obesity is linked with an increased risk for noncommunicable diseases including cardiovascular diseases, diabetes, musculoskeletal disorders, liver disease and some cancers (WHO, 2018). In 2013, WHO developed the "Global Action Plan for the Prevention and Control of Noncommunicable Diseases 2013-2020". This action plan lists nine global targets including "halt the rise in diabetes and obesity" and "a 10% relative reduction in prevalence of insufficient physical activity (PA)" (WHO, 2013).

1.1.6.1 The global prevalence of overweight and obesity

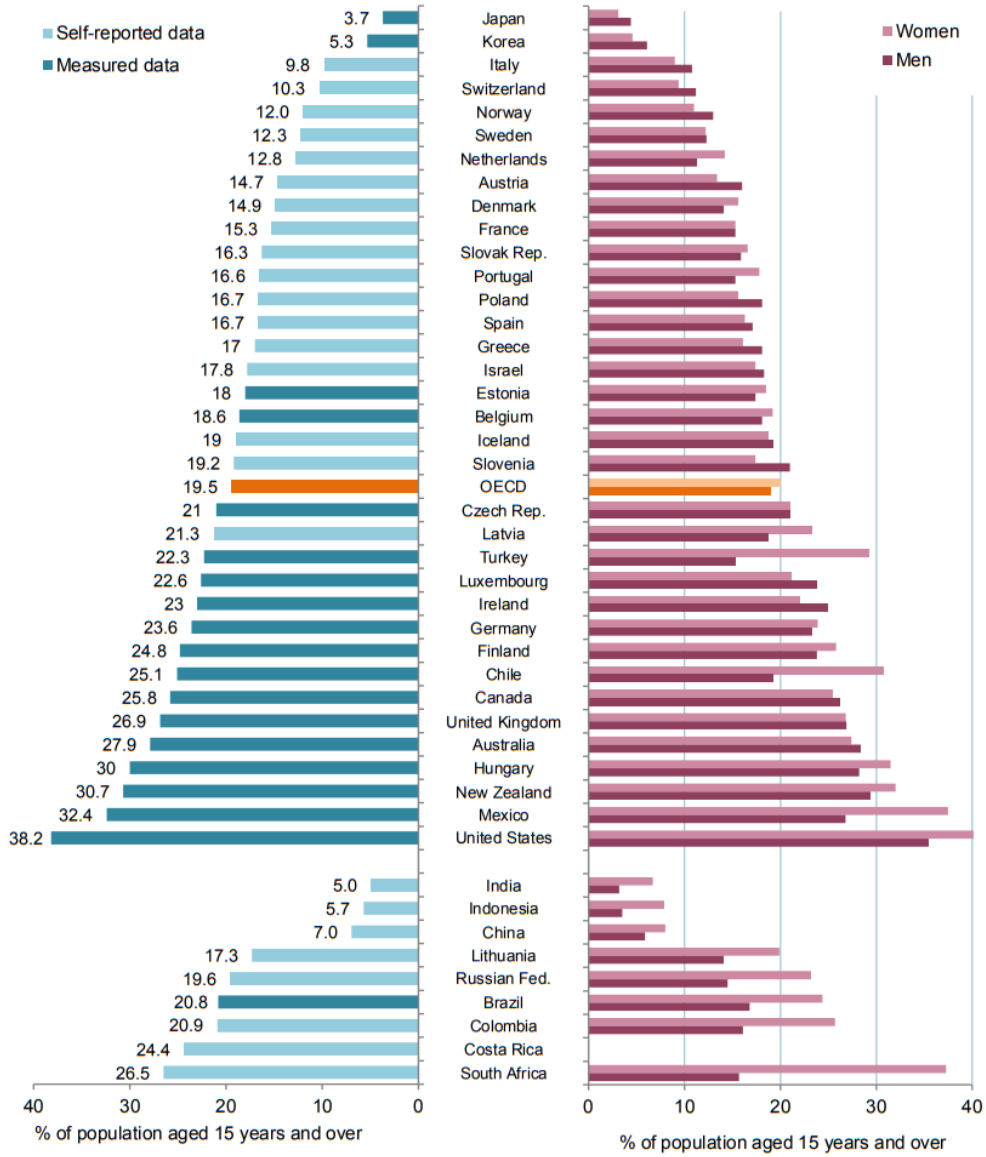
In 2016, the worldwide prevalence of overweight and obesity among women were 40% and 15% respectively. Although obesity was previously considered a

high-income country problem, it is now increasing in low- and middle-income countries (WHO, 2018).

In 2015, across The Organisation for Economic Co-operation and Development (OECD) countries, the highest prevalence of obesity is seen in Hungary, New Zealand, Mexico and the United States (more than 30%) and the lowest prevalence of obesity in Korea and Japan (less than 6%) (see figure 1.5) (OECD, 2017). During the last decade, whilst the prevalence of obesity has further increased in Canada, Mexico, Switzerland and the United States, it has remained at a similar level in England, Italy, Korea and Spain (ibid).

Between 1993 and 2016 in England, the prevalence of adults who were obese (BMI≥30 kg/m^2) increased from 15% to 26%. In 2016, 27% of women in England were classified as obese and 30% of women were classified as overweight (BMI between 25 and 29.9 kg/m^2) (Health and Social Care Information Centre, 2018).

Figure 1.5 Obesity among adults, 2015 or nearest year (OECD, 2017)

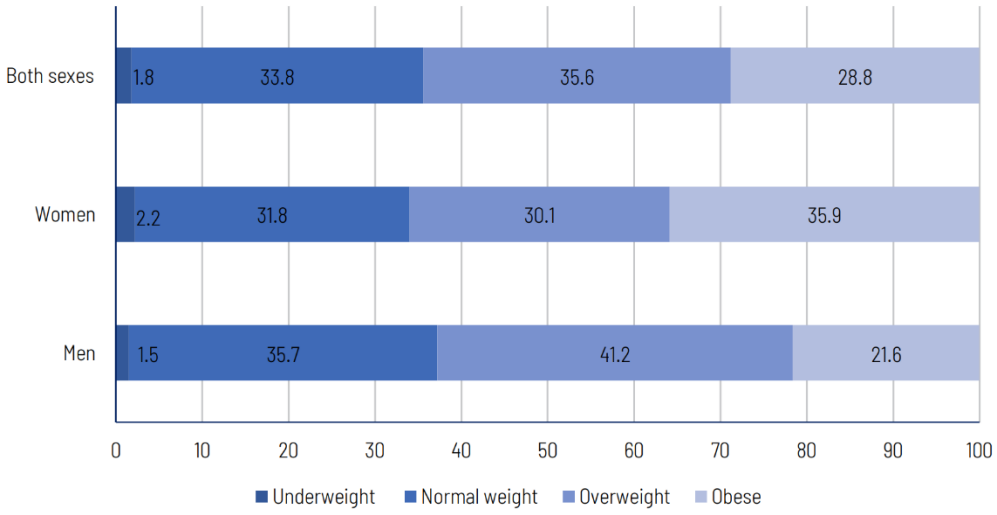


Note: India, Indonesia, China, Lithuania, Russian Federation, Brazil, Colombia, Costa Rica and South Africa were separated at the bottom of the diagram as these countries are not the members of the OECD.

1.1.6.1 The prevalence of overweight and obesity in Turkey

According to recent National Household Health Survey in Turkey (2017), the prevalence of overweight (BMI between 25 and 29.9 kg/m^2) and obesity (BMI≥ 30 kg/m^2) in adults aged fifteen and over were 35.6% and 28.8%, respectively. 41.2% of men and 30.1% of women were classified as overweight and 21.6% of men and 35.9% of women were classified as obese (WHO, 2018) (see figure 1.6). Whilst the prevalence of overweight in women in Turkey was similar to the prevalence of overweight in women in the UK, the prevalence of obesity in women in Turkey was greater than the prevalence of obesity in women from the UK.

Figure 1.6 Distribution of BMI category, by sex in Turkey (WHO, 2018)



Turkey's Demographic and Health Survey (2013), which is carried out every five years, showed that the obesity problem is getting greater among females. It was indicated that the prevalence of overweight in the 15-49 age group of women was found to be 33.4%, 34.2%, 34.4% and 55% and the prevalence of obesity was found to be 18.8%, 22.7%, 23.9% and 27% in 1998, 2003, 2008 and 2013 respectively. It was also reported that there is a positive correlation between mean BMI and the woman's age. Mean BMI increased from 22.5 kg/m^2 in the 15-19 age group to 30.7 kg/m^2 in the age group of 40-49 (Hacettepe University Institute of Population Studies, 2014).

According to the Ministry of Health of Turkey (2010), there was an inverse association between the family income status and the obesity prevalence. Whilst the obesity prevalence was 22.6% in regions with the lowest socio-economic status, in regions with high socio-economic status it was 17.6%.

Regarding profession, the highest obesity prevalence was in housewives (30.7%). Obesity was prevalent in 18.4% of retired people, 18.8% of unemployed people, 17.4% of workers, 14.9% of civil servants and 15.1% of the self-employed. Regarding education level, an inverse relationship between education level and obesity prevalence was observed. The obesity prevalence was 33.4% among people with no or incomplete primary education, 22.7% among primary school graduates, 8.7% among high school graduates and 10.0% among university graduates (Ministry of Health of Turkey, 2010).

1.1.7 Iron deficiency and obesity: the connection

Obesity is a major public health issue in both developed and developing countries. At the same time, ID is the most prevalent micronutrient deficiency globally (WHO, 2016). Both IDA and obesity represent malnutrition and result in a double health burden. The inverse relationship between iron status and obesity was first reported in 1962 (Wenzel *et al*., 1962).

There is a significant lower mean serum iron concentration in overweight individuals compared with normal-weight individuals (Cepeda-Lopez *et al*., 2010). Possible reasons for the high prevalence of ID among obese individuals include; 1) inadequate dietary iron intake due to poor food choices, 2) increased iron requirements because of increased blood volume, and 3) low-grade inflammation due to excess adiposity which influences circulating hepcidin levels and reduces iron absorption. These will be considered individually in the following paragraphs.

1.1.7.1 Inadequate dietary iron intake in overweight/obese individuals

Changes in the food supply towards to highly-processed, energy-dense and nutrient-poor products is closely related with increased global obesity prevalence (Crino *et al.*, 2015). The association between ultra-processed food consumption and higher risk of obesity was revealed by a number of studies (Mendonca *et al*., 2016; Canella *et al*., 2014 and Louzada *et al*., 2015). In a Canadian study, the relationship between consumption of ultra-processed foods and the nutrient profile of the diet in individuals (aged 2 years and above, n=33,694) was investigated and it was found that there was a significant positive

relationship between the dietary contribution of ultra-processed foods and the dietary content in carbohydrates, free sugars, total and saturated fats and energy density, and there was an inverse relationship with the dietary content in protein, fibre, many vitamins and minerals including iron (Moubarac *et al*., 2017). One of the possible explanations for the correlation between obesity and iron-deficiency anaemia may be poor dietary iron intake in overweight/obese individuals due to poor food choices.

In order to determine the energy intake, energy expenditure, and diet-composition of children and adolescents in Northern Greece, Hassapidou *et al*. (2006) conducted a study. The results of a three-day weighed dietary diary suggested that both overweight boys and girls had lower iron intakes than their non-overweight counterparts and both overweight and non-overweight adolescents had lower than recommended mean iron intakes (boys: 9.8±4.4 mg vs. 11.4±5 mg [overweight vs. non-overweight], girls: 8.1±4.8 mg vs. 9.4±4.2 mg [overweight vs. non-overweight] $p<0.05$). Also, overweight adolescents consumed more snacks and more sugar, jam and honey, and fewer legumes, vegetables and fruits than their non-overweight counterparts. However, these results were not consistent with a more recent study by Thongprasert *et al*. (2017) which compared adolescents (12-14 years) who were overweight/obese and their peers who were normal weight regarding intakes of dietary iron, and enhancers and inhibitors of iron absorption according to their parental education levels. It was found that dietary iron intake, which was determined via a 3-day prospective dietary record, and available iron (the proportion of iron available for intestinal absorption) were similar between adolescents who were overweight/obese and their peers who had normal weight. However, the intake

of iron, available iron, and haem- iron were significantly higher in adolescents whose mothers were less educated than their peers with mothers with a high-intermediate level of education among the group of adolescents who were overweight/obese.

Analysis of data from the 1999 Mexican Nutrition Survey, which included 1174 children (aged 5-12) and 621 non-pregnant women (aged 18-50) suggested that mean serum iron concentrations were significantly lower in obese women than in the normal-weight group (62.6±29.5 compared with 72.4±34.6 mg/dl; p=0.03). Whilst dietary energy and calcium intakes were significantly lower in obese women than in the normal-weight group, no differences were found in terms of dietary iron or ascorbic acid intakes between BMI groups. Their average iron intake was 44% of the Recommended Dietary Allowance (RDA) with no difference between normal-weight, overweight and obese groups (Cepeda-Lopez *et al*., 2011). However, these findings must be interpreted with caution because there may be under-reporting of dietary intake by women who are obese (Braam *et al*., 1998). It was also found that concentration of C-reactive protein (CRP) – the main acute phase protein and also a sensitive marker of systemic inflammation – were higher in obese women and children and were positively associated with BMI and negatively associated with iron status ($p < 0.05$) (Cepeda-Lopez *et al*., 2011). More recently, Suchdev et al. (2017) suggested that inflammation has confounding effects on iron indicators and this may lead to the incorrect diagnosis of malnutrition in individuals as well as the overestimation or underestimation of the prevalence of deficiency in a population. In terms of obesity, the need for well designed studies validating the different approaches of adjusting iron status (e.g. serum ferritin vs bone marrow

iron stores) and inclusion of biomarkers of inflammation was suggested (Suchdev et al., 2017).

These results are in line with those of some published studies (Tussing-Humphreys *et al*., 2010a; Moschonis *et al*., 2012; Knez *et al*., 2017) which confirmed that 'poor dietary iron intake' may not be sufficient to explain the underlying cause of the relation between impaired iron status and obesity.

In most developing countries, socioeconomic, cultural, or religious constraints may influence the consumption of flesh foods which are rich sources of haem iron. Instead, staple diets are predominantly plant-based which contain high levels of inhibitors of iron absorption such as dietary fibre, and phytate (Gibson *et al*., 2000). Therefore, fortification is a common public health intervention if the food to be fortified (e.g. wheat and maize flour and rice) is widely consumed in sufficient amounts by at-risk individuals (Pasricha *et al*., 2013). In economically-developed countries, besides mandatory fortification of staple foods such as wheat flour (began in 1953 in UK), market-driven fortifications are also available such as the voluntary fortification of breakfast cereals (Lim *et al*., 2013).

1.1.7.2 Increased iron requirements

Increasing body mass leads to a non-linear increased blood volume (Lemmens *et al*., 2006). Bertinado *et al*. (2014) examined the influences of diet-related obesity on iron status in rats that were categorised as obese or lean according to their percentage of body fat. The rats in both categories were given 5 mg/kg (low), 15 mg/kg (marginal), 35 mg/kg (normal) and 70 mg/kg (high) amounts of iron. The results showed that although hepcidin concentration was similar in all

rats, obese rats receiving low and marginal amounts of iron had lower serum iron than lean rats in the same diet group. It was concluded that an increase in body mass and blood volume in obesity leads to additional iron needs.

Cepeda-Lopez *et al.* (2018) investigated whether blood volume might explain the reduced serum iron in women who are overweight/obese. Sixty-four women of childbearing age were recruited and classified as normal weight (n=24), overweight (n=20) and obese (n=20). Dual energy X-ray absorptiometry was used to assess body composition and the carbon monoxide (CO)-rebreathing method was used to assess blood volume. It was found that there was a close correlation between blood volume and total lean mass ($r=0.83$, $p<0.001$), fat mass ($r=0.55$, $p<0.001$), waist circumference ($r=0.52$, $p<0.001$), height ($r=0.58$, $p<0.001$) and BMI ($r=0.50$, $p<0.001$). Also, a positive correlation was found between serum iron and Hb ($r=0.32$, $p=0.01$), body iron stores ($r=0.38$, $p=0.002$) and serum ferritin ($r=0.34$, $p=0.01$), and an adverse relationship was found between iron status and BMI ($r=-0.37$, $p=0.003$), weight ($r=-0.36$, $p=0.004$), fat mass ($r=-0.38$, $p=0.002$), alpha-1-acid glycoprotein ($r=-0.27$, $p=0.04$), and plasma volume ($r=-0.25$, $p=0.05$). Blood volume was also negatively associated with iron status, but the result was not statistically significant ($r=-0.20$, $p=0.11$).

The following equations to calculate blood volume in overweight/obese women were developed (Cepeda-Lopez *et al.*, 2018).

(1) Blood volume (ml)= 4698.8 * height (m) + 32.5 * weight (kg) - 5342.1 ($r^2=0.69$, $p<0.01$)

(2) Blood volume (ml)= 2686.7 * height (m) + 18.9 * weight (kg) + 66 * lean mass - 3932.3 ($r^2=0.76$, $p<0.01$)

This study suggested that hypoferremia is overestimated in populations with a high prevalence of obesity when using serum iron as an indicator after adjusting for the dilution effect of blood volume. However, more research on this topic needs to be undertaken before the need for an increase in the iron requirements of obese female individuals is more clearly understood.

1.1.7.3 Obesity-related inflammation and reduced iron absorption

Obesity is linked with low-grade inflammation and adipocytes have a crucial role in this relationship. Many inflammatory cytokines, including tumour necrosis factor-α (TNF-α), visfatin, interleukin-6 (IL-6) and anti-inflammatory adiponectin, are released by adipocytes (Wang & Nakayama, 2010). An association between higher concentrations of pro-inflammatory mediators and the pathogenesis of some obesity-related disorders including insulin resistance, type 2 diabetes mellitus, fatty liver disease and cardiovascular diseases has been proposed (Shoelson & Goldfine, 2009; Fuentes *et al*., 2013).

The first study to demonstrate the negative impact of adiposity on iron absorption was carried out by Zimmermann *et al*. (2008). In this study, women and children from transition countries, including Thailand, Morocco and India, were recruited to investigate the relationship between BMI and iron absorption. In the first experiment, healthy pre-menopausal Thai women (n=67) were recruited to consume iron-isotope-labelled test meals which consisted of foods typical of the Thai diet but containing approximately 4 mg of isotopically labelled fortification iron as [^{57}Fe/^{58}Fe]- ferrous sulphate. A first blood sample was collected prior to the meal to analyse haemoglobin, serum ferritin and CRP in order to assess inflammation. A second blood sample was collected for the

determination of isotope concentration and the calculation of iron absorption using mass spectrometry fourteen days after the test meal. It was reported that fractional iron absorption was negatively correlated with CRP and BMI ($p=0.030$). In their second experiment, responsiveness to iron fortification was evaluated in children and adolescents from Morocco and India aged 5-16 years ($n=1688$). In order to assess iron status, haemoglobin, serum ferritin and TfR were measured at baseline and following the 7–9-month fortification protocols. It was demonstrated that there was an inverse association between body iron (the serum ferritin/TfR ratio) and BMI Z-score ($p<0.001$). Also, in respect of iron fortification, an inverse relationship between BMI Z-score and a change in body iron was found.

These results are in agreement with the findings of a cross-sectional sample of women of child-bearing age in Chile studied by Mujica-Coopman *et al*. (2015) which assessed the association between BMI and both iron absorption and iron status. The results of the study suggested that iron absorption was significantly lower in obese women ($p< 0.05$) compared with overweight and normal-weight women. However, women with obesity had higher serum ferritin and haemoglobin ($p<0.05$) concentrations. Despite the existence of ID, serum ferritin is often elevated as a consequence of inflammation. A cross-sectional analysis of factors anticipated to influence serum iron by Yanoff *et al.* (2007) showed a significant increase in levels of both CRP and serum ferritin in the obese populations. According to McClung & Karl (2009) the hepcidin-inflammation connection could succinctly clarify the proposed mechanistic link between obesity and poor iron status (see Figure 1.7).

Figure 1.7 Proposed mechanistic link between obesity and poor iron status (McClung & Karl, 2009).



In order to explain the relationship between obesity and an alteration in iron homeostasis, a number of studies have focused on hepcidin. A study by Tussing-Humphreys *et al.* (2010a) was carried out on elevated systemic hepcidin and iron depletion in obese pre-menopausal females. Forty pre-menopausal women were recruited and classified into two groups as obese (n=20) and non-obese (n=20). The results of the determination of systemic hepcidin levels and their association with body mass revealed that women who are obese had significantly higher hepcidin levels compared with the non-obese women ($p<0.0001$) despite similar Hb levels. It was suggested that obese women have a hepcidin-induced iron deficiency in which dietary iron is inefficiently absorbed.

The link between hepcidin and obesity was also clearly supported by the findings of Bekri *et al*. (2006), who found that there was an increase in hepcidin expression in obesity, both at the messenger ribonucleic acid (mRNA) and the protein levels. Hepcidin expression was 150 times higher in the liver than in adipose tissue, but the latter contributed 60-70kg of body weight in these patients, which exceeds corresponding liver mass by approximately twenty times. Adipose tissue therefore makes a major contribution to circulating hepcidin concentrations in individuals who are overweight or obese independent

of diabetes and non-alcoholic steatohepatitis. Although there is a positive correlation between liver hepcidin expression and transferrin saturation (that is, those with greater iron concentrations have an appropriate feedback regulation to limit iron absorption and bioavailability by increasing hepatic hepcidin), no correlation was observed between hepcidin expression in subcutaneous adipose tissue and transferrin saturation ratio (Bekri *et al*., 2006).

Herter-Aeberli *et al.* (2015) assessed the risk of micronutrient deficiencies in relation to weight status in Indian women with a focus on iron but also including zinc and vitamins A and B and compared fractional iron absorption between obese and normal-weight women. It was found that overweight and obese middle-class women in India have an increased risk of developing iron deficiency compared with their normal-weight counterparts but are not at an elevated risk of deficiency of any of the other micronutrients analysed. Also, overweight/obese subjects had significantly higher CRP, serum ferritin, soluble transferrin receptor (sTfR) and hepcidin concentrations (analysed by competitive immunoassay) compared with normal-weight women ($p<0.05$). Increasing hepcidin secretion leads to reduced iron absorption in overweight/obese subjects. Using a stable iron isotope, it was further demonstrated that iron absorption was impaired in obese compared with normal-weight subjects. It was therefore recommended that the double burden of malnutrition should be considered in the planning of prevention strategies, especially in transition countries with emerging increasing prevalence of obesity.

Almost every study that has been carried out on ID in obesity found higher ferritin levels in people with obesity despite the existence of ID. Khan *et al*. (2016) explored whether ferritin is a marker of inflammation or ID in people who are overweight or obese. Similarly, a strong positive correlation with both BMI ($r=0.86$, $p<0.001$) and CRP ($r=0.87$, $p<0.001$) and strong negative correlation with Hb, iron, total iron binding capacity (TIBC), and transferrin saturation ($p<0.001$) were found. It was suggested that ferritin was a marker of inflammation rather than ID in people with overweight or obesity and ID can be truly predicted by complete iron profile including transferrin in this group of people.

1.1.7.4 The health consequences of a combination of obesity and iron deficiency anaemia

Obesity and IDA are two major public health issues independent of each other. Whilst obesity has harmful effects on health physically, psychologically and socially in all age groups, IDA gives rise to impaired cognitive development and poor growth in pre-school-aged children and to reduced work productivity and cognitive and behavioural problems in adults (Sachdev *et al*., 2005; Grandone *et al*., 2015; Murray-Kolb *et al*., 2007). Although no evidence for the health consequences of the combination of overweight and obesity with iron deficiency in adults was found, Grandone *et al*. (2015) reported a link between obesity, iron deficiency anaemia and cognitive dysfunction in children. In adults, this combination may result in cognitive impairment, poor exercise capacity, lack of motivation to lose weight and difficulty in weight loss independent of motivation. In order to gain a better understanding of the health consequences of the interaction of overweight and ID and to determine an intervention to treat

the issue, further quantitative and qualitative studies need to be carried out. Although quantitative studies have recognised the higher prevalence of ID in people with obesity, to date, no qualitative study has been performed to investigate the effect of ID on obesity or fatigue experience of people who are obese.

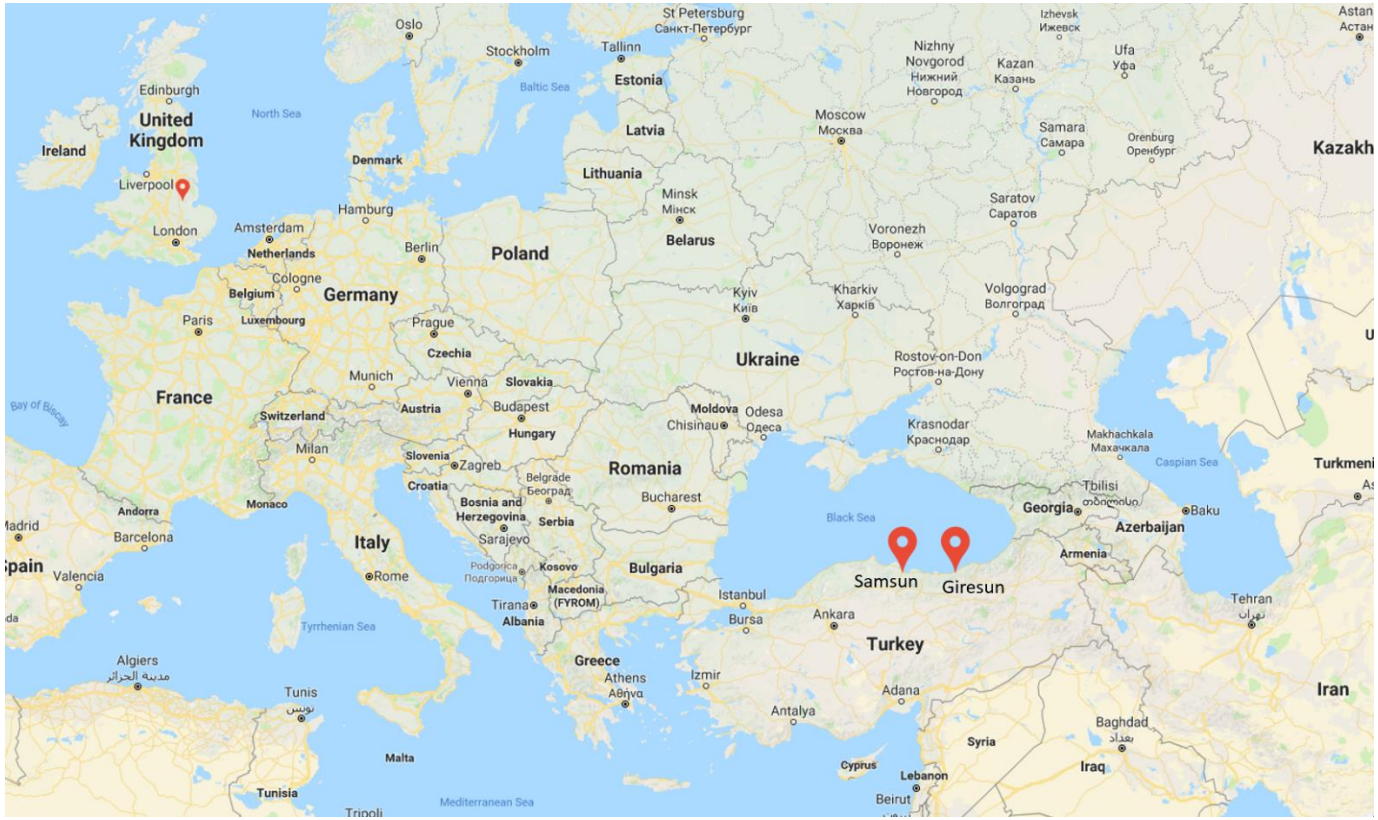
1.2 STUDY CONTEXT

1.2.1 Geographical context of Turkey

Turkey is an intercontinental Eurasian state, located mainly in Western Asia (97%) and Southeast Europe (3%) (Vinokurov & Libman, 2012). The country borders the Black sea, between Bulgaria and Georgia, and the Aegean Sea and Mediterranean Sea between Greece and Syria (see figure 1.8). Turkey occupies 786,562 sq km, of which 769,632 sq km is land and 13,930 sq km is water. While 27% of the total land is arable, 4% of the total land is permanent crops (CIA, 2018).

Turkey is the 18th most crowded country in the world with a population of 80,845,215. Almost a quarter (24.7%) of the population is between 0-14 years, 16% between 15-24 years, 43.2% between 25-54 years, 8.6% between 55-64 years and 7.5% 65 years and over. The median age is 30.5 years. The most major urban area of Turkey is Istanbul with a population of 14.75 million, and Ankara (the capital) follows it with a population of 4.92 million (CIA, 2018).

Figure 1.8 Location of Turkey on world map and Turkey's city map (the cities where the data collection took place are marked with a location icon).



1.2.2 Slimming World

Slimming World (SW) is a UK based weight management organisation, founded in 1969. Currently, there are over 19,000 groups held weekly around the UK and Ireland. Each week approximately 900,000 members attend the groups. The members are supported by a bi-monthly SW magazine and online via a members-only website (Slimming World, 2018).

The SW programme has been developed based on the most recent research on the multi-component approach compromises of three key principles nutrition, exercise and the psychology of behaviour change.

1.2.2.1 Food Optimising

Food Optimising is heathy eating plan based on three main components.

a) Free Foods: Members are encouraged to consume low energy dense and highly satiating foods to satisfy their appetite while reducing overall energy intake. These foods can be eaten without restriction including poultry, fish, lean meat, pasta, grains, vegetables and fruit.

b) Heathy Extras: Members are encouraged to include appropriate calcium and fibre-rich foods to achieve a good overall balance of nutrients.

c) Syns: Syns help members limit consumption of high energy dense and less satiating foods including saturated fats, alcohol and free sugars (Slimming World, 2018).

1.2.2.2 Body Magic

Body Magic is an activity programme that aims to help members increase their PA levels gradually. The programme supports behavioural change using stage-by-stage encouragements and rewards (Slimming World, 2018).

1.2.2.3 IMAGE (individual motivation and group experience) Therapy

This element aims to increase the confidence and self-esteem of the members by using group behavioural change techniques around eating and activity habits. SW's support system incorporates individual attention and group participation. This system encourages members to discuss their experiences, identify their own patterns of behaviour and discover new ways of overcoming barriers to change (Slimming World, 2018). SW uses elements of behavioural change techniques including Transactional Analysis, influence of Ego-States on behaviour and Motivational Interviewing to help individuals overcome their own personal barriers to change and to avoid relapse (Slimming World, 2019).

Transactional analysis is the model of self-concept based on the idea of three concurrent ego states: Ego-Child, Ego-Parent, Ego-Adult. The model was introduced by Berne (1958) who defined an ego state as "a consistent pattern of feeling and experience directly related to a corresponding consistent pattern of behaviour" (Berne, 1961). Saito *et al.* (2009) identified the psychological characteristics assessed by ego states in patients participated in a 6-month weight-loss program in a hospital obesity clinic (n=147). In this study transactional analysis therapy was used to improve patients' self-effectiveness and self-control in order to reduce psychological stress and to maintain the

weight loss. The psychosocial characteristics of participants were assessed pre- and post-intervention. It was showed that self-monitoring skill- and autonomy-related ego states of participants were increased during the weight-loss programme.

Motivational interviewing that is used by SW is an evidence-based approach designed to improve people's motivation for making desired changes in their lives and adherence to treatment (Miller & Rollnick 2012). In a systematic review (Armstrong *et al*., 2011), reviewed randomized controlled trials that investigated the effectiveness of motivational interviewing for reducing body mass, measured by change in body weight or BMI in adults who are overweight or obese. Meta-analysis of 11 studies in this review indicated that motivational interviewing was associated with a greater decrease in body mass compared to controls (Standardised mean difference = -0.51 [95% CI -1.04, 0.01]). In a more recent systematic review (Barnes & Ivezaj, 2015), motivational interviewing was examined for weight loss among adults within primary care settings. In this systematic review 24 randomised controlled trial were identified and 19 of those not included in the previous review (Armstrong *et al*., 2011). Whilst 9 studies reported positive effect of motivational interviewing on weight loss, 13 studies reported that patients receiving motivational interviewing achieved at least 5% loss of initial body weight. It was concluded that motivational interviewing is likely to support primary care patients for weight loss.

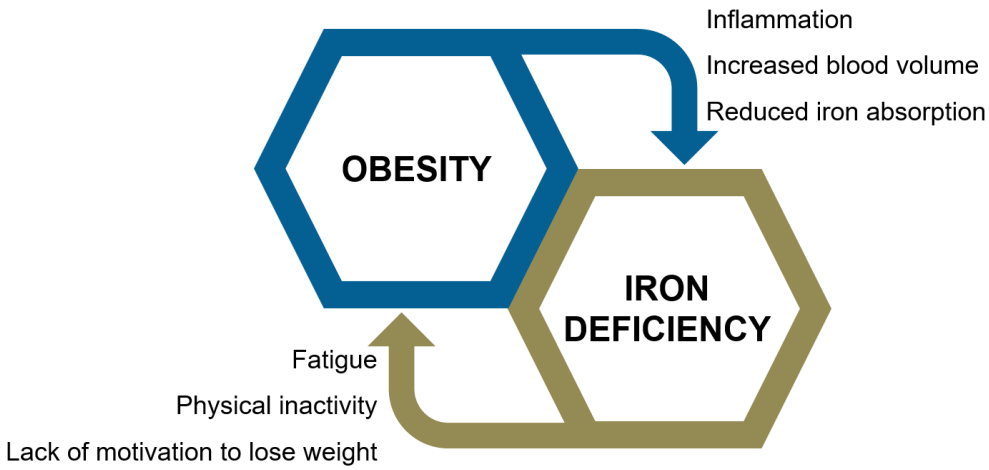
1.2.3 Study rationale

People are consuming more high-processed, energy-dense, and micronutrient-poor foods and drinks globally (Hawkes *et al*., 2017). At a population level, this

nutrition transition leads to a "triple burden" of malnutrition which includes three related but distinctly different problems: overt malnutrition, micronutrient deficiencies, and obesity (Pinstrup-Andersen, 2007). At the individual level, it is falsely assumed that people with obesity who have a high energy intake exceed all their nutritional requirements (Damms-Machado *et al*., 2012). Whilst the dietary intake of macronutrients may be greater, the intake of micronutrients such as iron may fall below recommendations. This situation leads to the "double burden" of malnutrition which is the co-occurrence of obesity and ID within the same individual. Despite the high prevalence of ID among people who are obese, much uncertainty still exists about the interaction between ID and adiposity (see figure 1.9), and the health consequences of a combination of obesity and ID. Furthermore, it is not clear what type of interventions could improve iron status in people with obesity.

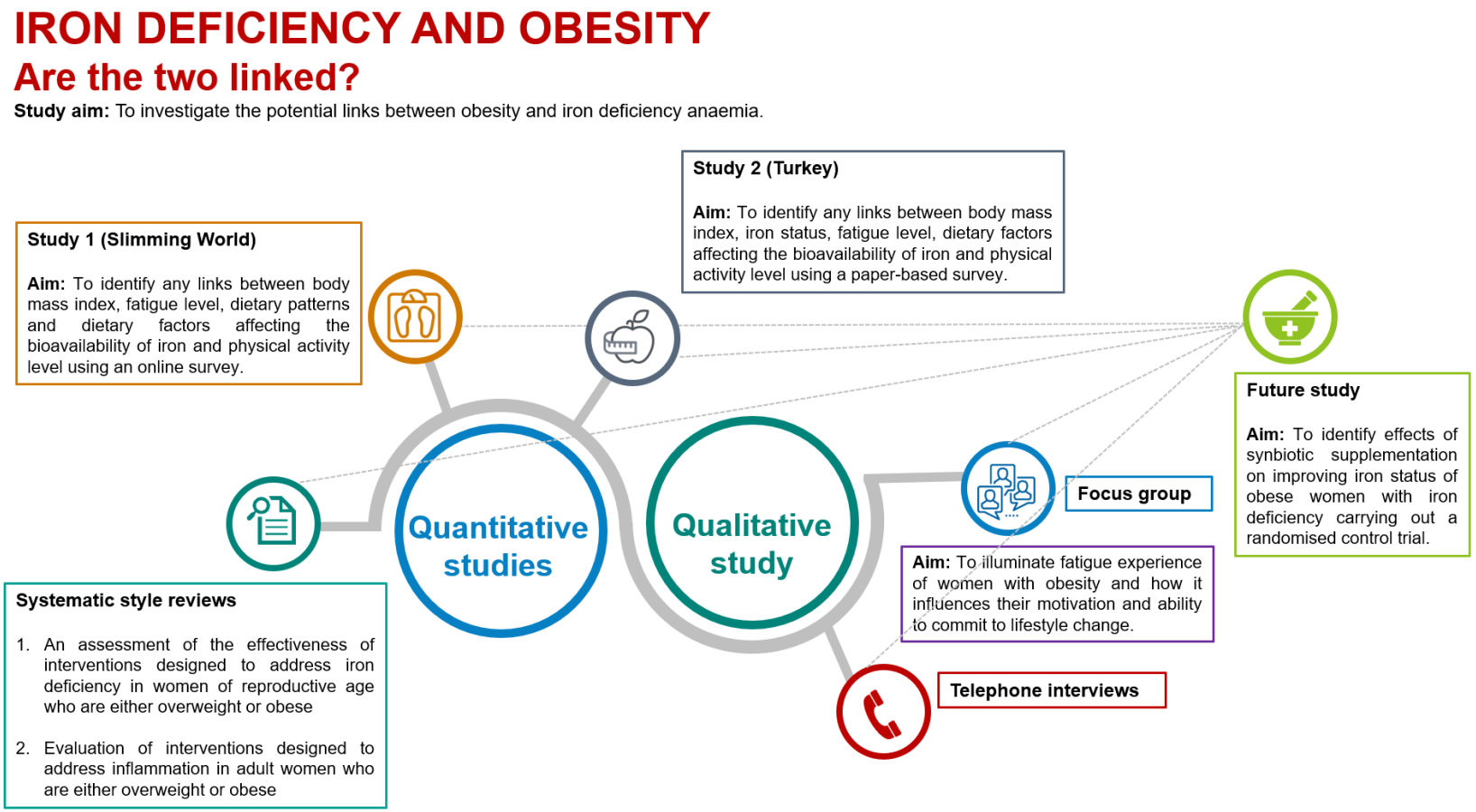
Fatigue, one of the most common symptoms of IDA, is also associated with higher BMI and percentage of body fat (Lim *et al*., 2005). This common symptom of both ID and obesity is one of the most widely reported barriers to lifestyle changes and PA (Timmerman, 1999; Kowal & Fortier, 2007). Even though behavioural changes and PA play crucial roles in providing sustainable weight management, there is limited scientific understanding of the effect of fatigue on a successful weight management programme and the effect of weight management programmes on level of fatigue. Currently, there is no qualitative data reporting the fatigue experience of people with obesity and how it influences their motivation and ability to commit to lifestyle change.

Figure 1.9 Potential interaction between obesity and ID



This thesis aims to investigate the research gaps which have been described above. The overall aim of this PhD is to both quantitatively and qualitatively investigate the potential links between obesity and ID, particularly in women of child-bearing age. To fulfil this aim, a mixed methodological approach was undertaken; two systematic style reviews exploring key questions, two quantitative studies and one qualitative study (see figure 1.10).

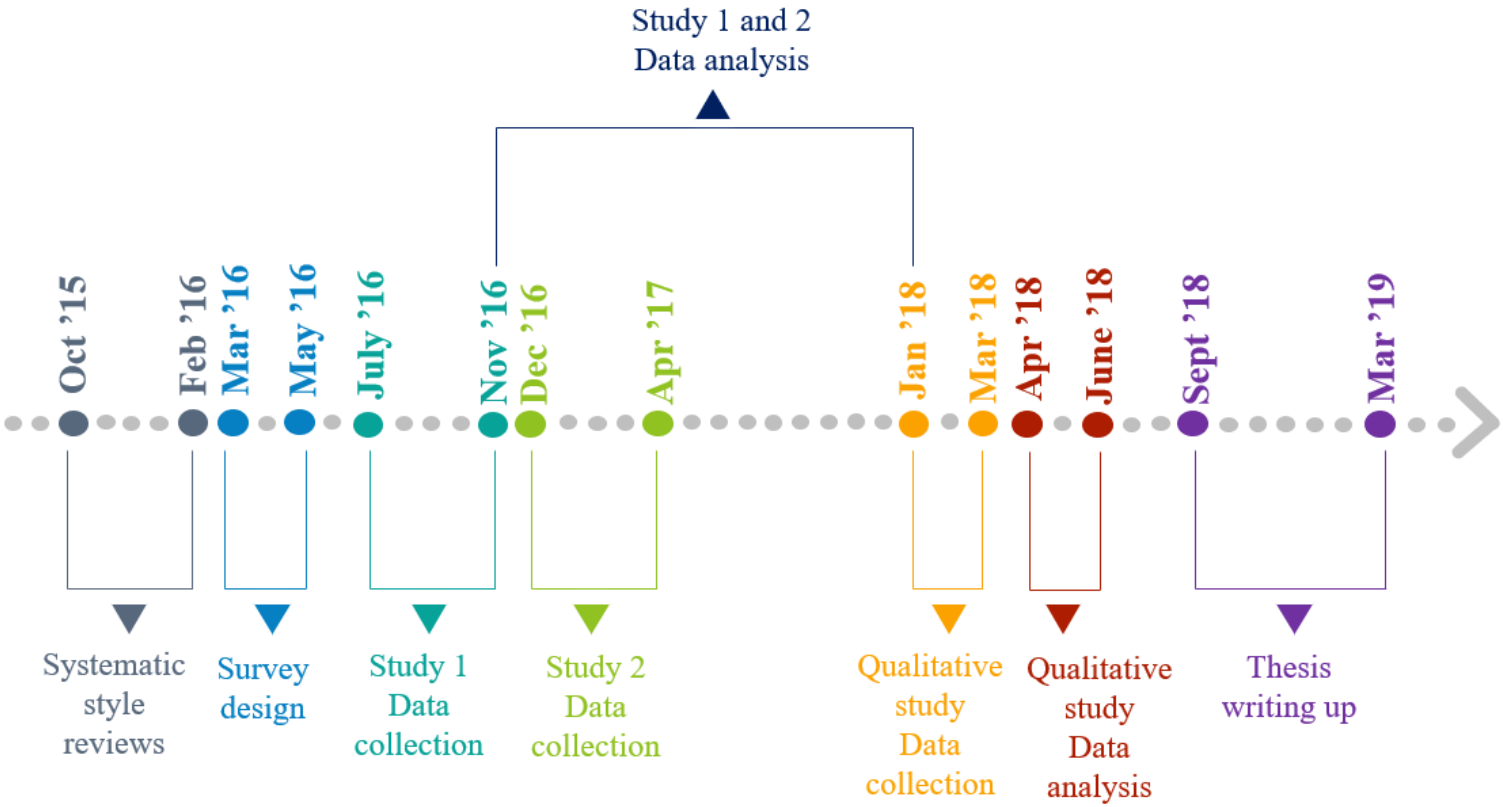
Figure 1.10 Thesis framework



Mixed method research studies benefit from the strengths of both quantitative and qualitative approaches and provides pragmatic advantages when exploring complex research questions (Fetters *et al*., 2013; McCusker & Gunaydin, 2015). Quantitative methodologies are used to obtain unbiased results that can be generalised to larger populations with the help of statistics (Glesne, 2015). Qualitative methodologies are used to investigate research questions to explore why or how people behave in certain ways as particularly relevant for health research that investigates human behaviour and relationships (Swift & Tischler, 2010).

The project was undertaken over a period of 36 months (see figure 1.11). During the first five months of the project, two systematic style reviews were conducted. Findings from the first systematic style review shaped the research questions of the second review. Results from the two systematic style reviews provided additional detail about the current gaps and knowledge and the research questions of main studies (two survey studies) were designed. The nature of the research questions drove the choice of mixed method research. After designing of study 1 based upon the research gaps, study 2 was modelled on study 1, but with cultural/practical adjustments on the survey and availability of iron-related blood test results. The quantitative studies provided detailed assessment of patterns of responses related to fatigue, dietary intakes and PA by statistical analysis. After conducting two large quantitative studies, a small qualitative study (due to the limited time available) was used to qualify the quantitative data by obtaining a deep understanding of fatigue experience of women who are obese.

Figure 1.11 Timeline of the current PhD research



1.2.4 Objectives of the study

The objectives of the PhD were the following:

1.2.4.1 Systematic style reviews

I. To assess the effectiveness of interventions designed to address iron deficiency in women of childbearing age who are either overweight or obese.

Research questions:

- What interventions have been developed to address ID in women of reproductive age who are either overweight or obese?
- What are the effects of these intervention on iron status?
- What are the effects of interventions that are not primarily designed to improve iron level, but assessed iron related parameters on iron status?

II. To explore the effectiveness of interventions designed to reduce inflammation in adult women who are either overweight or obese.

Research questions:

- What interventions have been developed to reduce inflammation in adult women who are either overweight or obese?
- What are the effects of these interventions on biomarkers of inflammation?

1.2.4.2 Study I

I. To determine mean BMI in a population of SW members before they started following Food Optimising (T0), whilst they are still a new member (around 4 weeks after joining) (T1) and 12 weeks later (T2) and identify BMI change over these time periods.

Research questions:

- What is the mean BMI of women who attended SW at T0, T1 and T2?
- Are there any significant changes in women's BMI over the three time points (T0, T1 and T2)?

II. To determine fatigue levels in women at T0, T1 and T2 and identify changes over these time periods.

Research questions:

- What is the mean Global Fatigue Index (GFI) of women who attended SW at T0, T1 and T2?
- Are there any significant changes in women's GFI over the three time points (T0, T1 and T2)?

III. To determine physical activity levels in women at T0, T1 and T2 and identify changes over these time periods.

Research questions:

- What is the mean total physical activity Metabolic Equivalent of Task (MET)-minutes/week of women at T0, T1 and T2?

- Are there any significant changes in women's total physical activity (PA) over the three time points (T0, T1 and T2)?

IV. To explore associations between weight change, fatigue level, and PA level in women at T0, T1 and T2.

Research questions:

- Is there a relationship between percentage weight change and baseline BMI, fatigue level, and PA level?

- Do baseline BMI and decrease in fatigue level predict weight loss?

- Is there any effect of the change in fatigue level on weight loss and PA level?

V. To identify the most frequently consumed food groupings in women at T0, T1 and T2.

Research question:

- What are the 35 most frequently food groupings at T0, T1 and T2?

VI. To determine dietary habits in women at T0, T1 and T2.

Research questions:

- What are the prominent dietary habits of women at T0, T1 and T2?

- Are there any changes in the dietary habits over the three time points?

VII. To determine iron-related dietary patterns of women who attended SW at T0 and T1.

Research question:

- What are the iron-related dietary patterns of women at T0 and T1?

VIII. To explore associations between BMI, level of fatigue and the dietary patterns at T0 and T1.

Research question:

- Is there any relationship between the identified dietary patterns and BMI and presence of fatigue?

1.2.4.3 Study 2

I. To determine mean BMI in Turkish women who consulted a registered dietitian in Turkey starting a healthy eating plan (T0) and 12 weeks after starting the plan (T1) and identify BMI change over these time periods.

Research questions:

- What is the mean BMI of women at T0 and T1?
- Are there any significant changes in women's BMI from T0 to T1?

II. To determine fatigue levels in women at T0 and T1 and identify changes over these time periods.

Research questions:

- What is the mean GFI of the women at T0 and T1?
- Are there any significant changes in women's GFI from T0 to T1?

III. To determine PA levels in women at T0 and T1 and identify changes over these time periods.

Research questions:

- What is the mean total PA (MET)-minutes/week of women at T0 and T1?
- Are there any significant changes in women's total PA from T0 to T1?

IV. To explore iron status of women at baseline.

Research questions:

- What are the mean values of iron-related blood parameters including haemoglobin, haematocrit and ferritin?
- What is the prevalence of IDA and low-iron store status?

V. To determine association of iron status with BMI and level of fatigue.

Research question:

- Is there any significant relationship between iron status, BMI and fatigue level?

VI. To identify the most frequently consumed food groupings in women at T0 and T1.

Research question:

- What are the 35 most frequently food groupings at T0 and T1?

VII. To determine dietary habits in women at T0 and T1.

Research questions:

- What are the prominent dietary habits of women at T0 and T1?
- Are there any changes in the dietary habits from T0 to T1?

VIII. To compare all available findings from UK and Turkey.

1.2.4.4 Qualitative Study

I. To determine fatigue experience in a population of Slimming World members.

II. To determine how fatigue influences their motivation and ability to commit to lifestyle change.

III. To determine how dietary changes affects their fatigue level.

IV. To determine how weight loss affects their fatigue level.

V. To explore their beliefs about how reducing fatigue levels may further improve their quality of life.

CHAPTER 2: SYSTEMATIC STYLE REVIEWS

This chapter presents two systematic style reviews. Both reviews followed the systematic review process, but paper identification was by the principal investigator, checked by both supervisors, rather than two or more independent researchers. Hence these reviews have been called- systematic style, rather than systematic reviews. The first review assessed the effectiveness of interventions designed to address iron deficiency in women of reproductive age, who are either overweight or obese. Obesity is associated with low-grade inflammation and increased hepcidin circulation reducing iron absorption, hence the second review evaluates interventions designed to address inflammation in women who are either overweight or obese.

2.1 THE EFFECTIVENESS OF INTERVENTIONS DESIGNED TO ADDRESS IRON DEFICIENCY IN WOMEN OF REPRODUCTIVE AGE WHO ARE EITHER OVERWEIGHT OR OBESE – A SYSTEMATIC STYLE REVIEW

2.1.1 Introduction

A strong association between low iron status and adiposity is confirmed from various studies (Cepeda-Lopez *et al*., 2010; Herter-Aeberli *et al*., 2015; Khan *et al.,* 2016). The combination of obesity and ID in an individual may trigger cognitive impairment, poor exercise capacity, lack of motivation to lose weight and difficulty in weight loss independent of motivation. Despite the importance of ID in people who are obese, no review investigating the outcomes of interventions designed to address ID in obesity was found (by searching the Database of Abstracts of Reviews of Effects (DARE) and the Cochrane

Database of Systematic Reviews (CDSR)). Therefore, the current review of literature was undertaken to address three questions:

1) What interventions have been developed to address ID in women of reproductive age who are either overweight or obese?

2) What are the effects of these intervention on iron status?

3) What are the effects of interventions that are not primarily designed to improve iron level, but assessed iron related parameters on iron status?

2.1.2 Subject and Methods

2.1.2.1 Search strategy and identification of studies

A systematic search of electronic databases was carried out in November 2016 to identify potentially relevant studies. The database search included PubMed, Google Scholar and Web of Science. For each online database investigation, the following keywords and a combination of these were used to develop a robust reference list: “iron”, “iron status”, “intervention”, “weight”, “obese”, “obesity”, “overweight”, “women”, and “female”. Also, the bibliographies of previous reviews were hand-searched to ensure relevant studies were not missed.

The assessment of trial eligibility was done by screening the title initially. After selecting relevant studies by title screening, the abstract was read. Full text of the studies that could not be excluded was reviewed and the assessment of study eligibility was done.

2.1.2.2 Inclusion and exclusion criteria

Papers published from January 1996 to December 2016 and presented in English were included. Studies were selected based on pre-determined inclusion and exclusion criteria. Inclusion criteria was framed in terms of participants, interventions, outcomes and study design.

Participants: non-pregnant women of child-bearing age (19-49 years) who are either overweight or obese.

Interventions: any intervention given for the improvement of iron status. Studies that included surgical interventions (e.g. bariatric surgery) were excluded.

Outcomes: studies where at least one standard assessment measure of iron such as haemoglobin, haematocrit and plasma iron were included as either primary or secondary outcome data.

Study design: randomised controlled trials and controlled trials. Case studies and case reports were excluded because these study designs have high bias risks.

2.1.2.3 Data extraction

Relevant information about study characteristics and findings from the included studies were obtained with a standardised data extraction form which was tailored to meet the review aim. Extracted data are presented in table 2.1 with included information; first author, publication year, country, participants, number, mean age, mean BMI, intervention, duration of the intervention, and effect/conclusion.

2.1.2.4 Quality assessment

Downs and Black's (1998) check list (see appendix L) was used to assess the methodological quality of the studies. This checklist was selected because it was designed to evaluate both quantitative randomised and non-randomised trials. Also, the tool was found to be easy to use and one of the best evaluation systems available, with well validated numerical rating scales (yes/no/unable to determine answers and clear descriptions of how to score items) in a review of 60 research design methodology evaluation systems (Deeks et al., 2003). There are 27 items pertaining to the quality of data reported, external validity, bias, confounding variables and power. The scoring of item 27 was modified (0 = insufficient power (or no information available); 1 = a power analysis was conducted) instead of the six-point scale used in the original version (rating from 0 to 5 based on the size of smallest intervention group). Therefore, the highest possible score for the checklist was 28 rather than 32. Total scores were evaluated based on the corresponding quality levels given by Hooper et al. (2008): excellent (26-28); good (20-25); fair (15-19); and poor (≤14). Whilst the quality level of two studies was found as fair, four studies were scored as good (see table 2.2).

Table 2.2 Modified Downs and Black checklist for the assessment of the methodological quality of studies in the first systematic style review

	Beard, 1997	Cepeda-Lopez, 2015	Cheng, 2013	Kretsch, 1998	Rodríguez-Rodríguez, 2007	Volpe, 2001
1. Aims	1	1	1	1	1	1
2. Outcomes	1	1	1	1	1	1
3. Participants characteristics	1	1	1	1	1	1
4. Clear intervention	1	1	1	1	1	1
5. Confounders	0	2	2	0	2	1
6. Findings	0	1	1	1	1	1
7. Random variability	1	1	1	1	1	1
8. Adverse outcomes	1	0	1	1	0	1
9. Patients lost to FU	1	0	0	1	1	1
10. Actual p value	0	1	1	1	0	0
11. Representative sample	1	0	0	1	1	0
12. Representative participants	0	0	0	0	1	0
13. Facilities	1	1	0	0	0	0
14. Blind participants	1	0	1	0	0	1
15. Blind assessor	1	0	0	0	0	1
16. Data dredging	1	1	1	1	1	1
17. FU length	1	1	1	1	1	1
18. Statistics	1	1	1	1	1	1
19. Treatment compliance	0	1	1	0	1	0
20. Outcome measures	1	1	1	1	1	1
21. Comparable groups	1	0	0	0	1	1
22. Comparable time frame	1	1	0	1	1	1
23. Randomised	0	1	1	0	1	1
24. Concealed randomisation	0	0	0	0	0	1
25. Adjustment for confounders	0	1	1	1	0	1
26. Accounting for FU loss	1	1	1	1	1	1
27. Power	0	1	1	0	0	1
Total	**18**	**20**	**20**	**17**	**20**	**22**

Yes=1, no=0, unable to determine=0 or yes=2, partially=1, no=0

2.1.3 Results

The initial literature review identified 296 papers for screening. Forty-seven articles were retrieved after screening titles and abstracts, and then assessed against the inclusion/exclusion criteria. Forty-two were excluded: three did not meet the language criterion; eight did not meet participants' age, gender and BMI criterion; seven did not include explicit outcomes and twenty-four did not meet the criterion for type of study. One study was identified from searching through reference lists. Consequently, six articles met the eligibility criteria for the review (see figure 2.1).

Figure 2.1 Flow chart of study selection process for the first systematic style review

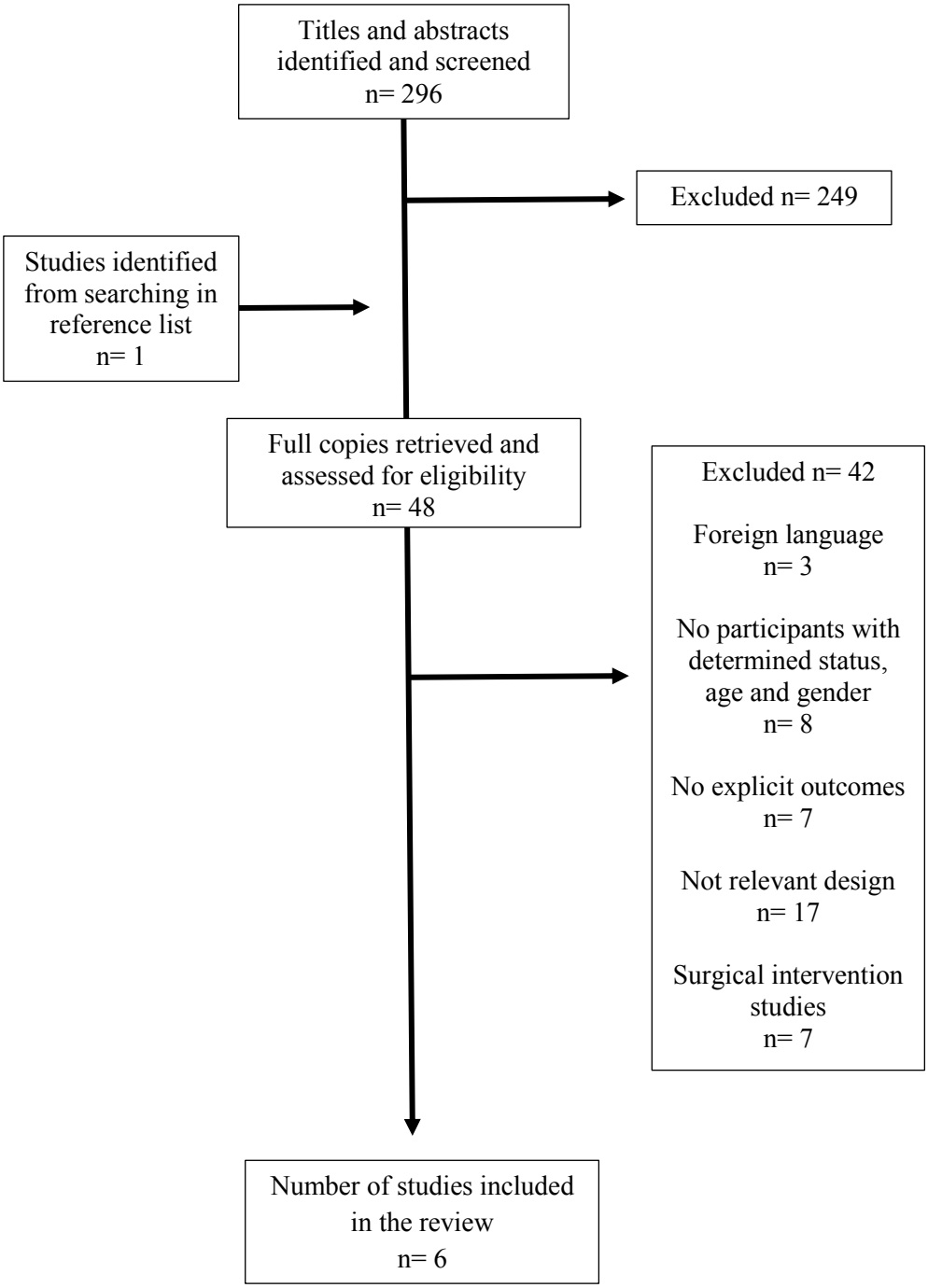


Table 2.1 Summary of the trials included in the review to assess the effectiveness of interventions which may improve iron status in women who are overweight or obese.

First author, year, country	Participants				Intervention		Effect/Conclusion
	Inclusion criteria	Number	Mean age (years)	Mean BMI (baseline) (kg/m^2)	Type	Duration	
Beard, 1997, USA	Free-living women who are obese	14	41	102.6 ± 14.9*	Lowest very low energy diet (420 kcal)	20 weeks	An increase in transferrin saturation values >16% ($p<0.001$) in middle very low energy diet + Fe group by week 20.
		17	39	101.1 ± 17.0*	Middle very low energy diet + 27 mg Fe/d (660 kcal)		
		12	46	100.5 ± 19.6*	Highest very low energy diet (800 kcal)		
Cepeda-Lopez, 2015, Switzerland	Healthy, nonanemic, women (normal weight, overweight and obese)	24	23	21.9 ± 1.9	**Day 1:** First labelled test meal without ascorbic acid (5 mg ^{57}Fe) **Day 2:** Second labelled test meal with ascorbic acid (5 mg ^{58}Fe+ ascorbic acid at a molar ratio of AA: iron of 2:1) **Day 16:** Blood sample	16 days	Median percentage increases in iron absorption for -AA to +AA meals were 56% in the normal weight group ($p<0.001$) and 28% in overweight and obese group ($p= 0.006$).
		19	26	27.3 ± 1.5			
		19	27	32.8 ± 2.8			

First author, year, country	Participants				Intervention		Effect/Conclusion
	Inclusion criteria	Number	Mean age (years)	Mean BMI (baseline) (kg/m^2)	Type	Duration	
Cheng, 2013, Australia	Young women who are overweight and obese	24	22	34.3 ± 3.5	Hypocaloric diet with higher-protein	6 months	HP diet showed higher median ferritin (52.0 vs 39.0 µg/L; p=0.021) and lower median soluble transferrin receptor-ferritin index (sTfR-F; 0.89 vs 1.05; p=0.024) at 12 months. Impact of ≥10% weight loss on iron was more apparent in LP diet who exhibited > mean serum iron (20.0 vs 13.5 µmol/L; p=0.002), transferrin saturation (29.8% vs 19.4%; p=0.001) and lower sTfR (1.24 vs 1.92 mg/L; p=0.034) at 12 months.
		20	22	32.2 ± 3.6	Hypocaloric diet with lower-protein		
		21	22	34.6 ± 3.4	Hypocaloric diet with higher-protein	12 months	
		15	22	32.2 ± 3.6	Hypocaloric diet with lower-protein		

First author, year, country	Participants				Intervention		Effect/Conclusion
	Inclusion criteria	Number	Mean age (years)	Mean BMI (baseline) (kg/m^2)	Type	Duration	
Kretsch, 1998, USA	Healthy, premenopausal, obese women	14	25-42	31.5	Baseline	3 weeks	A significant reduction in haemoglobin, haematocrit, and red blood cell count occurred across the study ($p<0.05$).
					50% caloric restriction	15 weeks	
					Weight stabilisation	3 weeks	
Rodríguez-Rodríguez, 2007, Spain	Healthy, young women	29	28	27.6 ± 2.6	**Diet C**: Weight control measures based on increasing the proportional consumption of cereals	6 weeks	There was an improvement in Hb levels at 6 weeks in women who initially had the poorest iron status (Hb< 12.6 g/L) and followed the Diet C.
		28		28.4 ± 3.4	**Diet V:** Weight control measures based on increasing the proportional consumption of greens and vegetables		

First author, year, country	Participants				Intervention		Effect/Conclusion
	Inclusion criteria	Number	Mean age (years)	Mean BMI (baseline) (kg/m^2)	Type	Duration	
Volpe, 2001, USA	Pre-menopausal, sedentary females	22	43	32.5 ± 4.3	Gel capsules containing 400 µg of chromium picolinate per day + moderately intense exercise program	12 weeks	Exercise training significantly reduced TIBC by 3% for all subjects combined (p = 0.001).
		22	43	33.2 ± 4.2	Placebo capsules (no chromium) + moderately intense exercise program		

*Mean weight was reported when mean BMI was not available

2.1.3.1 Details of included studies

Beard *et al*., (1997) assessed the effects of very low energy diets (VLEDs), over a 12 week period, on indicators of iron and thyroid status where the iron content of the diet varied from 18 to 27 mg/d and the energy intake from 420 to 800 kcal/d. Forty-three free-living women participated and were assigned to three dietary groups: lowest VLED (n=14), middle VLED (n=17), and highest VLED (n=12) after completing 1 week of a 1200 kcal/d prescribed diet consumed at home. The lowest VLED (420 kcal/d) included 70g protein, 30g carbohydrate and 2g fat and the RDA of all micronutrients (including 18 mg iron/d). The middle VLED (660 kcal/d) included 90g protein, 30g carbohydrate and 20g fat and the RDA of all micronutrients with a total of 27 mg iron/d and 180 mg ascorbic acid/d. The highest VLED (800 kcal/d) included 70g protein, 100g carbohydrate and 13g fat and the RDA of all micronutrients, including 18 mg iron/d as ferrous sulphate. All diets were consumed as liquid-diet formulations five times per day over the 12 weeks. Assessment of venous blood samples indicated that the middle VLED group had significantly higher transferrin saturation (>16% increase, $p<0.001$) values than those in the other groups at the end of the protocol (week 20) despite the similar baseline values. Plasma ferritin concentration decreased significantly in lowest VLED and highest VLED diet group ($p<0.001$) by week 20. The greatest weight loss was also observed in the middle VLED group with an inverse correlation between decline in plasma ferritin level and body weight loss ($r=-0.36$).

Cepeda-Lopez *et al*., (2015) compared iron absorption from a standardised test meal with ascorbic acid and a standardised test meal without ascorbic acid in

women who have normal weight (BMI between 18.5-24.9 kg/m^2, n=24) and with women who are overweight (BMI between 25-29.9 kg/m^2, n=19) and obese (BMI between 30-39.9 kg/m^2, n=19). The test meal (white rolls with butter and honey) without ascorbic acid (labelled with 5 mg ^{57}Fe) was consumed by the participants on day 1. The test meal with ascorbic acid (labelled with 5 mg ^{58}Fe and ascorbic acid) was consumed on the second day. A fasting blood sample was collected to assess iron status and level of inflammation on day 16. The study showed that fractional iron absorption from the meal without ascorbic acid was 19.0% in the normal-weight group and 12.9% in the overweight/obese group. Whilst iron absorption increased to 29.5% from the meal with ascorbic acid in the normal-weight group, it increased to only 16.6% in the overweight/obese group. It was also found that there was a negative correlation between BMI and serum iron (r=-0.37, p=0.003) and TIBC (r=-0.34, p=0.007), and a positive correlation between BMI and some inflammatory markers including CRP (r=0.39, p=0.002) and IL-6 (r=0.38, p=0.003).

Cheng *et al.* (2013) conducted a 12-month single-blind randomised controlled study to compare the effect of two hypocaloric diets with different contents of protein, haem iron and zinc on iron, zinc and inflammatory status of young women who are overweight and obese (BMI ≥27.5 kg/m^2). The primary outcome measures were considered in relation to the degrees of weight loss (<5% and ≥10%). The participants assigned to either higher-protein (HP) diet (n=36) or lower-protein (LP) diet (n=35). Whilst the HP diet compromised of 32% protein; 41% carbohydrate; 5% fat; 11.7 mg zinc; 12.2 mg iron (1.90 mg haem iron), the LP diet compromised of 20% protein; 58% carbohydrate; 21% fat; 7.60 mg zinc; 9.90 mg iron (0.40 mg haem iron). Results reported in this

study referred to six (n=44) and 12-month (n=36) completers only. A significant weight loss was achieved by the participants in both groups over 6 and 12 months with the HP diet leading to more weight loss (approximately double) than the LP diet. In terms of iron status, the participants who followed the HP diet had higher median ferritin (52.0 vs 39.0 μg/L, p=0.021) levels at 12 months and lower median soluble transferrin receptor/ferritin index, an indicator of iron status in inflammation (0.89 vs 1.05, p=0.024), than those who followed LP diet during the same period. The participants in the LP diet group who lost ≥10% of their initial weight had higher serum iron (p=0.002), transferrin saturation (p=0.001) and lower soluble transferrin receptor (p=0.034) than those in the same diet group who lost <5% their initial weight at 12 months.

Kretsch *et al.* (1998) carried out a longitudinal study to determine the relationship between cognitive function and iron status in women who are obese (body weights between 130-150% of ideal body weight) and desire to lose weight (n=14). The study included three periods: baseline (3 weeks), 50% energy restriction (15 weeks) and weight stabilisation (3 weeks). Formulated meals were provided for the participants to take home except for one meal per day that was consumed at the research centre. During the baseline and weight stabilisation periods, sufficient energy for body weight maintenance was provided by the formulated diet. During the 50% energy restriction period, 50% of the baseline calories was provided. Iron intake was maintained from food sources (50%) and an oral supplement (50%) (approximately between 15-18mg/d in total). By the end of caloric restriction weeks, the participants lost an average of 12.3±5.5 kg of body weight (as 10.4 kg fat). Assessment of haematological parameters which were measured during all three periods

showed that there was a significant decrease in Hb, haematocrit, red blood cells and TIBC despite the provision of sufficient dietary iron.

Rodríguez-Rodríguez *et al*. (2007) conducted a study to compare the effects of two different six-week weight control programs (Diet C and Diet V) on iron status in a group of women who were overweight or mildly obese (BMI between 24-35 kg/m^2). Diet C encouraged subjects to decrease the intake of energy-rich foods and increase cereal consumption (especially breakfast cereal) (n=29). Diet V also restricted the intake of energy-rich foods but encouraged the participants to consume more greens and vegetables (minimum 3 times per day) (n=28). Both diets were slightly hypocaloric (20% less than the theoretical energy requirements of the subjects). At the end of the intervention, both diets were effective in supporting weight loss (in a similar amount). Assessment of the food and drink records showed that participants in both groups increased the intake of total and non-haem iron but decreased the intake of haem iron. In terms of iron related parameters, it was reported that there was a reduction in transferrin levels in both groups at 6 weeks but there was an improvement in Hb levels at 6 weeks in women who initially had the poorest iron status (Hb< 12.6 g/L) and followed Diet C.

Volpe *et al*., (2001) undertook a double-blind, placebo-controlled study to investigate the effects of chromium supplementation on several factors including body composition and iron status in women who were moderately obese (BMI between 27-41 kg/m^2) and participated in a 12-week exercise programme. Although previous researchers established that chromium may improve glucose tolerance (Anderson *et al*., 1991) and result in weight loss (Grant *et al*., 1997)

in some individuals, results were ambiguous. In this study, the subjects in chromium supplementation group received gel capsules containing 400 µg chromium picolinate per day (n=22) and the control group received identical looking placebo capsules (n=22). All participants engaged in a 12-week supervised moderately-intense exercise programme (two days per week, one hour each session). At the end of the study, no significant changes in body weight and BMI in either group was reported. For iron status there was a slight decrease in serum iron level, transferrin saturation and serum ferritin concentration in both groups over time. TIBC significantly decreased by 3% over time in both groups in response to exercise training (p=0.0011).

2.1.3.2 Key findings

This review revealed that no intervention study has been specifically designed to primarily address ID in women of reproductive age who are either overweight or obese. Five trials in this review investigated iron status of women of reproductive age who are either overweight or obese although the studies were not designed to improve iron level (Beard *et al*., 1997; Cheng *et al*., 2013; Kretsch *et al*., 1998; Rodriguez-Rodriguez *et al*., 2007; Volpe *et al*., 2001). Two studies assessed the influences of hypocaloric diets (Beard *et al*., 1997; Kretsch *et al*., 1998), one study examined the effects of vegetable-rich and cereal-rich diets (Rodriguez-Rodriguez *et al*., 2007), and one study compared the effects of higher-protein and lower-protein diets on some iron related parameters (Cheng *et al*., 2013). One study compared the effects of ascorbic acid on iron absorption in women who have normal weight and overweight/obese (Cepeda-Lopez *et al*., 2015).

Four studies in this review found positive effects of the interventions on iron status. An additional small amount of dietary iron (9 mg/d) above the RDA with very low energy diets (Beard *et al*., 1997); high-protein diet and weight loss more than 10% of the initial weight (Cheng *et al*., 2013); breakfast cereals (Rodriguez-Rodriguez *et al*., 2007); and exercise training (Volpe *et al*., 2001) were found to be effective in improving some diagnostic biomarkers of IDA. However, one study (Kretsch *et al*., 1998) suggested that hypocaloric diets, even when adequate dietary iron is available, impair iron status and could lead to ID in women who are obese. It was reported that decrease in Hb may be associated with subclinical eating disorder. The only study that assessed iron absorption rather than iron-related parameters showed that in women who are overweight and obese, the enhancing effect of ascorbic acid is approximately half that of women who are a healthy weight.

2.1.4 Discussion

This review aimed to investigate and evaluate interventions designed to address ID in women in childbearing age who are either overweight or obese. The key finding to emerge from this review is that although intervention trials which assess iron status are available, to date, no study has been undertaken where the primary aim is to improve iron status of women who are obese. The majority of studies in this review reported beneficial impacts of the interventions on some iron related parameters in women who are overweight or obese. The effective interventions reported in this study included additional dietary iron in very low energy diet, higher protein and weight loss, breakfast cereals and exercise training.

Weight loss was a common theme in three out of four interventions that found positive effects on iron status (Beard *et al*., 1997; Cheng *et al*., 2013; Rodriguez-Rodriguez *et al*., 2007). This result corroborates the findings of previous reports of the relationships between adiposity, inflammation, hepcidin circulation and iron absorption. Tussing-Humphreys *et al*. (2010b) investigated the influence of weight reduction on iron status, inflammation and serum hepcidin in premenopausal females who are obese (n=20) 6 months after restrictive bariatric surgery. The study showed that there were significant decreases in BMI (47.56 vs. 39.55 kg/m^2), CRP (10.83 vs. 5.71 mg/l) and serum hepcidin (111.25 vs. 31.35 ng/ml) and significant improvements in the levels of Hb (12.10 vs. 13.30 g/dl) and haematocrit (36.58 vs. 38.78%) between baseline and 6 months after surgery. It was suggested that dietary iron intake did not change after the surgery. The study concluded that there was a correlation between weight loss and decreased serum hepcidin and inflammation in premenopausal women who are obese. However, these findings do not support one of the studies in this review (Kretsch *et al*., 1998) that found that energy restriction can impair iron status in women with obesity despite the presence of adequate dietary iron and achievement of weight loss. This rather contradictory result may be due to modulation in protein metabolism which may affect iron storage and release in periods of energy deprivation.

All trials included in this review assessed serum ferritin levels and the findings were inconsistent. In this review, low serum ferritin level was not considered as a marker of impaired iron status because of its interaction with inflammation in people who are obese. Findings from Beard *et al*. (1997) and Cepeda-Lopez *et al*. (2015) appear to support this. Whilst Beard et al. (1997) reported an inverse

correlation between body weight loss and ferritin concentration (r=-0.36), Cepeda-Lopez *et al*. (2015) reported a positive correlation of serum ferritin with percentage body fat (r=0.26) and serum hepcidin (r=0.59). Furthermore, three studies (Beard *et al*., 1997; Rodriguez-Rodriguez *et al*., 2007; Volpe *et al*., 2001) reported a decrease in serum ferritin levels by the end of the interventions probably because of decreased inflammation related to either weight loss (Beard *et al*., 1997; Rodriguez-Rodriguez *et al*., 2007) or increased physical activity (Volpe *et al*., 2001). However, more research on this topic needs to be undertaken before the association between ferritin and obesity-related inflammation is more clearly understood. Although studies have recognised higher prevalence of ID among women with obesity, research has yet to systematically investigate the effect of interventions on iron status.

There was a major source of uncertainty in terms of the comparison between studies due to the different methodologies used to assess iron status including Hb, soluble transferrin receptor and TIBC. Whilst some of the markers used to assess iron status were affected by contaminant inflammation such as serum ferritin, some of the markers were affected/limited from a lack of assay standardisation such as soluble transferrin receptor (Pfeiffer & Looker, 2017). Therefore, this study had the challenge of appropriate interpretation of iron status assessment both within an indvidual study (pre- and post-intervention) and between studies.

The strengths of this study included the comprehensive search and provision of a novel approach to this area. Although selection and quality assessment of trials was performed by one researcher (HKK), other assessors (AA and FM – PhD

supervisors) verified the appropriateness of the paper selection and whether they met the inclusion criteria.

The questions raised by this study are “to what extent could weight loss be effective in improving iron status in premenopausal women who are obese” and “what are the interventions designed to address inflammation over and above weight loss in women who are obese and are these interventions effective to improve iron status”. The second part of this chapter will offer some important insights into to the effects of intervention on inflammation in women who are obese.

2.2 INTERVENTIONS DESIGNED TO ADDRESS INFLAMMATION IN ADULT WOMEN WHO ARE EITHER OVERWEIGHT OR OBESE – A SYSTEMATIC STYLE REVIEW

2.2.1 Introduction

Obesity is linked with low-grade inflammation. Over the last 10 years approximately, evidence has emerged that adipocytes play a crucial role in releasing many inflammatory cytokines such as TNF-α, visfatin, IL-6 and anti-inflammatory adiponectin (Wang & Nakayama, 2010). In addition, adipose tissue has been identified as a major contributing factor to the increase of hepcidin expression which inhibits iron absorption (Bekri *et al*., 2006). This is one of the possible reasons for the high prevalence of ID among obese individuals. Therefore, interventions to reduce inflammation could improve iron status in people who are obese, by decreasing hepcidin expression, and improving iron absorption. Before investigating the effects of strategies to reduce inflammation on iron status, there is a need to further understand the effectiveness of interventions designed to address inflammation in adults who are obese.

There are systematic reviews of interventions which have investigated the effects of weight loss (Selvin *et al*., 2007), exercise (Simpson & Singh, 2008), consumption of dairy products (Labonte *et al*., 2013), flaxseed and its derivatives (Ren *et al*., 2016), and plant-based diets (Eichelmann *et al*., 2016) on biomarkers of inflammation in an overweight/obese population or healthy and overweight/obese population. However, to date, there is no systematic

review specifically looking at the effectiveness of anti-inflammatory interventions in overweight or obese populations of women. Therefore, this study aims to contribute to this growing area of research by exploring the effectiveness of interventions designed to reduce inflammation in adult women who are either overweight or obese, by addressing two questions:

1) What interventions have been developed to reduce inflammation in adult women who are either overweight or obese?

2) What are the effects of these interventions on biomarkers of inflammation?

2.2.2 Subjects and Methods

2.2.2.1 Search strategy and identification of studies

A systematic style approach was used for the literature review. The search was conducted in January 2016. Papers published in last 10 years (from January 2006 to December 2016) in English were included. The database searched included PubMed, Google Scholar and Web of Science. The search strategy included search terms such as: “inflammation”, “low-grade inflammation”, “CRP”, “obese”, “obesity”, “overweight”, “women”, and “female”. These key words were combined with ‘AND’ or ‘OR’ for the specific searches. Also, the bibliographies of previous reviews were interrogated manually. The method for the assessment of trial eligibility was the same as the previous review from title screening to full text reading (see section 2.1.2.1).

2.2.2.2 Inclusion and exclusion criteria

Studies were selected based on pre-determined inclusion and exclusion criteria. Inclusion criteria were framed in terms of participants, interventions, outcomes and study design.

Participants: studies involving adult women (over 18 years) who are either overweight or obese.

Interventions: any intervention aiming to reduce inflammation.

Outcomes: studies where at least one standard biomarker of inflammation, such as TNF-α and IL-6 which are expressed by adipose tissue and secreted into the systemic circulation (Calder *et al.,* 2011), and CRP which is the best indicator to assess inflammation and subject to intensive research in number of studies globally (Shrivastava *et al.,* 2015) , was reported.

Study design: randomised controlled trials and controlled trials. Case studies and case reports were excluded.

2.2.2.3 Data extraction

A standardised data extraction form was created and necessary information about study characteristics and findings from the included studies were obtained. Extracted data are presented in table 2.3 and included information on; first author, publication year, country, inclusion criteria, number, mean age, mean BMI, intervention type, duration of the intervention, results, and effect/conclusion.

2.2.2.4 Quality assessment

Study quality assessment was carried out using Downs and Black's (1998) check list and it was suggested that the quality level of seven studies were fair and three of those were good (see table 2.4)

Table 2.4 Modified Downs and Black checklist for the assessment of the methodological quality of studies in the second systematic style review

	Alves, 2013	Basu, 2009	Cheng, 2013	Crane, 2011	Faraj, 2011	Feng, 2013	Ghavipour, 2013	Kazemzadeh, 2014	Rosado, 2011	Ryan, 2014
1. Aims	1	1	1	1	1	1	1	1	1	1
2. Outcomes	1	1	1	1	1	1	1	1	1	1
3. Participants characteristics	1	1	1	1	1	1	1	1	1	1
4. Clear intervention	1	1	1	1	1	1	1	1	1	1
5. Confounders	1	1	2	1	1	0	0	0	0	2
6. Findings	1	1	1	0	1	1	1	1	1	1
7. Random variability	1	1	1	1	1	1	1	1	1	1
8. Adverse outcomes	1	0	1	0	0	0	0	0	1	0
9. Patients lost to FU	0	1	0	0	1	1	1	1	1	1
10. Actual p value	1	0	1	1	1	0	1	1	1	0
11. Representative sample	0	0	0	1	1	1	0	0	1	0
12. Representative participants	0	0	0	1	1	1	0	0	1	0
13. Facilities	0	0	0	0	0	0	0	0	0	0
14. Blind participants	1	0	1	0	0	1	0	0	1	0
15. Blind assessor	0	0	0	0	0	1	0	0	1	0
16. Data dredging	1	1	1	1	1	1	1	1	1	1
17. FU length	1	1	1	1	1	1	0	1	1	1
18. Statistics	1	1	1	1	1	1	1	1	1	1
19. Treatment compliance	0	1	1	0	0	0	0	0	0	0
20. Outcome measures	1	1	1	1	1	1	1	1	1	1
21. Comparable groups	1	1	0	1	1	1	1	1	1	1
22. Comparable time frame	0	1	0	0	1	1	1	1	1	1
23. Randomised	1	0	1	1	0	1	1	1	1	0
24. Concealed randomisation	0	0	0	0	0	1	0	0	1	0
25. Adjustment for confounders	0	0	1	0	0	0	0	0	0	0
26. Accounting for FU loss	1	1	1	1	1	1	1	1	1	1
27. Power	1	1	1	0	0	1	1	1	1	0
Total	**18**	**17**	**20**	**16**	**18**	**21**	**16**	**17**	**23**	**16**

Yes=1, no=0, unable to determine=0 or yes=2, partially=1, no=0

2.2.3 Results

Five hundred and eighty-four articles were identified in the initial literature search. Sixty-eight articles were retrieved after screening titles and abstracts, and then assessed against the inclusion/exclusion criteria. Sixty-one were excluded: six did not meet the language criterion; thirty-two did not meet participants' age, gender, and BMI criterion; eleven did not include explicit outcomes and twelve did not meet the criterion for type of study. Three articles were identified from searching the reference lists. Consequently, ten articles met the eligibility criteria for the review (see figure 2.2).

Figure 2.2 Flow chart of study selection process of the second systematic style review

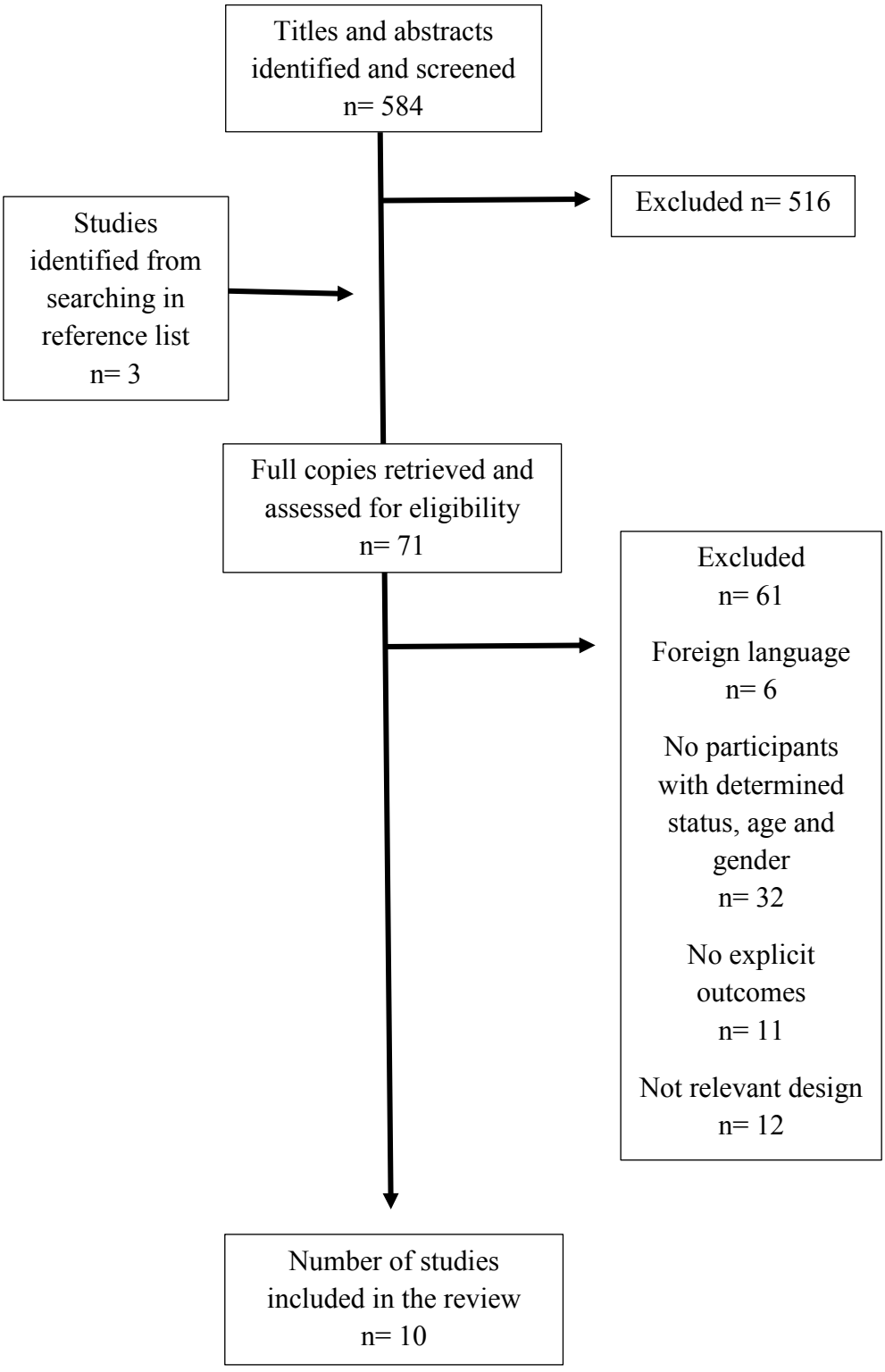


Table 2.3 Summary of the trials included in the review to address inflammation in adult women who are either overweight or obese

First author, year, country	Participants				Intervention		Results			Effect on inflammation
	Inclusion criteria	No	Mean age	Mean BMI (baseline)	Type	Dur	Change in BMI (kg/m^2)	Change in waist circum. (cm)	Change in hs-CRP (mg/l)	
Alves, 2013, Brazil	Women who are obese	11	35	28.7 ± 2.9	**CR:** Hypocaloric diet and control drink consumption	5 weeks	28.7 ± 2.9 vs 28.4 ± 3.0 p=0.02	96.0 ± 7.5 vs 94.9 ± 7.8 p=0.05	4.30 ± 5.6 vs 4.90 ± 6.2 p=0.53	There was no significant change in hs-CRP amounts, ferritin levels and total leukocyte count.
		11			**CRHR:** Hypocaloric diet and consumption of drink containing ‘‘Human Ration’’		28.8 ± 2.9 vs 28.5 ± 3.0 p=0.01	97.1 ± 7.6 vs 94.6 ± 7.6 p=0.0003	4.70 ± 5.7 vs 3.80 ± 3.2 p=0.97	
Basu, 2009, USA	Women with metabolic syndrome	16	52	38.6 ± 2.3	Consumption of two cups of the strawberry drink (25 g freeze-dried strawberry powder in water) daily.	4 weeks	38.6 ± 2.3 vs 38.8 ± 2.3 p≥0.05	109.2 ± 3.8 vs 111.8 ± 3.8 p≥0.05	9.5 ± 0.3 vs 9.3 ± 0.5 p≥0.05	No effects were noted on markers of inflammation including CRP and adiponectin.

First author, year, country	Participants				Intervention		Results			Effect on inflammation
	Inclusion criteria	No	Mean age	Mean BMI (baseline)	Type	Dur	Change in BMI (kg/m^2)	Change in waist circum. (cm)	Change in hs-CRP (mg/l)	
Cheng, 2013, Australia	Young women who are overweight and obese	24	22	34.3 ± 3.5	Hypocaloric diet with higher-protein	6 months	9.3 ± 8.9% (weight loss) p<0.001	-7.8 ± 1.3 p=0.030	3.55 vs. 2.36 p=N/A	There were reductions in CRP levels in both intervention groups, but changes were insignificant.
		20	22	32.2 ± 3.6	Hypocaloric diet with lower-protein		5.1 ± 6.0% (weight loss) p=0.001	-3.7 ± 1.0 p=0.030	4.63 vs. 2.65 p=N/A	
		21	22	34.6 ± 3.4	Hypocaloric diet with higher-protein	12 months	9.8 ± 13.0% (weight loss) p=0.003	-7.9 ± 1.8 p=0.036	3.51 vs. 2.22 p=N/A	
		15	22	32.2 ± 3.6	Hypocaloric diet with lower-protein		4.6 ± 7.2% (weight loss) p=0.027	-2.4 ± 0.8 p=0.036	4.75 vs. 3.95 p=N/A	

First author, year, country	Participants				Intervention		Results			Effect on inflammation
	Inclusion criteria	No	Mean age	Mean BMI (baseline)	Type	Dur	Change in BMI (kg/m^2)	Change in waist circum. (cm)	Change in hs-CRP (mg/l)	
Crane, 2011, USA	Postmenopausal women who are overweight /obese	49	59	32.0 ± 5.0	2 servings of vegetables daily (130g) 5 servings of vegetables daily (287g) 10 servings of vegetables daily (614g)	3 weeks	N/A	N/A	No significant reduction (data not shown)	No significant change in oxidative stress and inflammation in overweigh, postmenopausal women.
Faraj, 2011, Canada	Postmenopausal women who are overweight /obese	56	58	32.3 ± 4.5	Hypocaloric dietary intervention (aim: 10% weight loss)	6 months	32.3 ± 4.5 vs 30.1 ± 4.2 $p<0.0001$	N/A	3.1 ± 2.3 vs 2.5 ± 2.1 $p<0.05$	There was a significant reduction in inflammation.

First author, year, country	Participants				Intervention		Results			Effect on inflammation
	Inclusion criteria	No	Mean age	Mean BMI (baseline)	Type	Dur	Change in BMI (kg/m^2)	Change in waist circum. (cm)	Change in hs-CRP (mg/l)	
Feng, 2013, China	Women with obesity and metabolic syndrome	45	47	30.7 ± 2.9	**Placebo group:** Four tablets of placebo daily	12 weeks	30.7 ± 2.9 vs 30.9 ± 2.4 p≥0.05	96.7 ± 6.2 vs 97.0 ± 6.0 p≥0.05	3.9 ± 2.5 vs 3.6 ± 2.7 p≥0.05	Histidine supplementation significantly decreased serum inflammatory cytokines (TNF-α, and IL-6).
		47	45	30.9 ± 2.6	**Histidine group:** 4 g histidine daily		30.9 ± 2.6 vs 30.0 ± 2.1 p<0.05	96.1 ± 5.2 vs 93.4 ± 5.4 p<0.05	4.0 ± 2.2 vs 2.7 ± 2.3 p≥0.05	
Ghavipour, 2013, Iran	Women who are overweight and obese	51	23	28.2 ± 0.3	**Control group:** Usual diet with water	20 days	N/A	N/A	2.8 ± 1.9 p≥0.05	Tomato juice reduces inflammation in overweight and obese females (significant decrease in IL-8 and TNF- α)
		53	23	28.3 ± 0.3	**Intervention group:** 330 ml tomato juice daily		N/A	N/A	5.0 ± 3.9 p≥0.05	

First author, year, country	Participants				Intervention			Results			Effect on inflammation
	Inclusion criteria	No	Mean age	Mean BMI (baseline)	Type		Dur	Change in BMI (kg/m^2)	Change in waist circum. (cm)	Change in hs-CRP (mg/l)	
Kazemzadeh, 2014, Iran	Non-menopausal women who are overweight or obese	20	33	29.4 ± 4	First intervention period	**Group 1:** Treatment with brown rice diet	Week 0-6	30.7 ± 3.5 vs. 29.6 ± 3.7 p=N/A	96 ± 9.3 vs. 89 ± 8.1 p=N/A	2.0 ± 1.3 vs. 1.9 ± 1.9 p=N/A	In the brown rice diet, inflammatory marker (hs-CRP) level decreased significantly in comparison with white rice diet.
		18				**Group 2:** Treatment with white rice diet		28.2 ± 3.5 vs. 27.6 ± 3.8 p=N/A	91 ± 10.5 vs. 88 ± 9.9 p=N/A	1.4 ± 0.7 vs. 2.0 ± 1.4 p=N/A	
		20			Second intervention period	**Group 1:** Treatment with white rice diet	Week 8-14	29.6 ± 3.6 vs. 29.4 ± 3.8 p=N/A	89 ± 8.1 vs. 87 ± 8.8 p=N/A	1.9 ± 1.6 vs. 2.5 ± 2.6 p=N/A	
		15				**Group 2:** Treatment with white rice diet		27.5 ± 3.7 vs. 26.9 ± 3.8 p=N/A	87 ± 9.1 vs. 82 ± 7.0 p=N/A	1.5 ± 1.2 vs. 0.9 ± 1.1 p=N/A	

First author, year, country	Participants				Intervention		Results			Effect on inflammation
	Inclusion criteria	No	Mean age	Mean BMI (baseline)	Type	Dur	Change in BMI (kg/m^2)	Change in waist circum. (cm)	Change in hs-CRP (mg/l)	
Rosado, 2011, Mexico	Women with obesity	46	35	34.8 ± 3.4	Energy-restricted diet + 250 ml of low-fat milk 3 times/day	16 weeks	34.9 vs 33.2 p<0.05	105.8 vs 96.1 p<0.05	0.2 (change) p≥0.05	Intake of low-fat milk with added micronutrients increases the effectiveness of an energy-restricted diet to treat obesity but had no effect on inflammation (CRP level).
		46	34	35.3 ± 5.5	Energy-restricted diet + 250 ml of low-fat milk with micronutrients three times/day		35.1 vs 32.7 p<0.05	107.5 vs 95.9 p<0.05	-0.7 (change) p≥0.05	
		47	34	34.8 ± 3.8	Energy-restricted diet + No milk		34.8 vs 32.3 p<0.05	106.2 vs 96.5 p<0.05	-1.1 (change) p≥0.05	

First author, year, country	Participants				Intervention		Results			Effect on inflammation
	Inclusion criteria	No	Mean age	Mean BMI (baseline)	Type	Dur	Change in BMI (kg/m^2)	Change in waist circum. (cm)	Change in hs-CRP (mg/l)	
Ryan, 2014, USA	Postmenopausal women who are overweight /obese and sedentary	37	60	32 ± 1	**AEX+WL:** Hypocaloric diet (-300 to -500 kcal/day) + aerobic exercise	6 months	32 ± 1 vs 30 ± 1 p<0.001	96.3 ± 2.4 vs 91.8 ± 2.5 p<0.001	-29% p<0.001	Combined weight loss and aerobic exercise led to decrease inflammation (CRP level).
		40	61	33 ± 1	**WL:** Hypocaloric diet (-300 to -500 kcal/day)		33 ± 1 vs 30 ± 1 p<0.001	96.7 ± 1.6 vs 91.0 ± 1.7 p<0.001	-21% p<0.05	

*Number- only participants who completed the intervention study

P values for within group differences (compared with baseline values)

N/A, not available

2.2.3.1 Details of included studies

Alves *et al*. (2013) performed a randomised controlled crossover study to investigate the efficacy of a new nutritional supplement (Human Ration) to reduce metabolic risk parameters in obese women (percentage body fat > 33%, n=22). The nutritional supplement consisted of a mixture of fourteen ingredients, including wheat fibre, defatted soy flour, triturated flaxseed, brown rice flour, rolled oats, white corn meal, triturated sesame cream, brown sugar, and wheat germ. Participants were allocated into two groups as CRHR and CR, with the CRHR group consuming a drink with Human Ration added with a hypocaloric diet (15% energy restriction of the estimated energy requirement), whereas the CR group received a control drink without Human Ration with a hypocaloric diet. After 6 weeks (5 weeks of intervention and 1 week of washout), the second part of the study started in which the participants in the CRHR group in the first period received treatment CR and the participants in the CR group in the first period received treatment CRHR. The mean value of high-sensitivity C-reactive protein (hs-CRP) represented subclinical inflammatory disease (4.80±5.7 mg/l) at baseline. At the end of the intervention, there was a significant weight loss (CR: -0.74±1.3 kg, p=0.01; CRHR: -0.77±1.3 kg, p=0.02) in the participants in both groups. Regarding inflammation, no significant differences in serum hs-CRP, ferritin, total leukocyte count, triglycerides and total antioxidant capacity was reported in any intervention. However, there was a significant association of the changes in hs-CRP with the changes in trunk fat (r=0.45, p=0.04) and android fat (r =0.47, p=0.03) in the control group.

Basu *et al.* (2009) assessed the effects of freeze-dried strawberry powder on fasting lipids and biomarkers of oxidative stress and inflammation in women with three features of the metabolic syndrome (BMI >30 kg/m^2, n=16). The participants were asked to consume two cups of a drink containing 25 g freeze-dried strawberry powder in one cup of water, with one teaspoon of artificial sweetener, and one teaspoon of vanilla essence per day for four weeks. The results of fasting blood indicated that there were no significant changes in hs-CRP (9.5±0.3 vs 9.3±0.5) and adiponectin concentration (9.7±1.0 vs 11.2±1.3) between baseline and end of the intervention.

Cheng *et al.* (2013) conducted a 12-month single-blind randomised controlled study to compare the effect of two hypocaloric diets with different contents of protein, haem iron and zinc on iron, zinc and inflammatory status of young women who were overweight and obese (BMI ≥ 27.5 kg/m^2, n = 71). Details of the study were discussed in the first part of this chapter (see "details of included studies" of the first review). In terms of inflammation, the study found that there were reductions in CRP levels in both intervention groups, with the high protein diet decreasing CRP more than the low protein diet (p = 0.085 at 12 months).

Crane *et al.* (2011) performed a randomised controlled crossover trial to test if the consumption of more vegetables is linked with a decrease in oxidative stress and inflammation in postmenopausal women who are overweight and obese (BMI between 25–45 kg/m^2, n=49). The participants completed a four-week low fruit/vegetable washout period and were randomly assigned to one of three vegetable intervention groups: 2 (130 g), 5 (287 g), and 10 (614 g) servings of vegetables daily. After completing the intervention (3 weeks), there was a

second washout period (4 weeks), after which the participants were randomised into the three intervention groups. Vegetables used in this intervention study were baby leaf lettuce green mix, tomatoes, red bell peppers, and baby carrots. In terms of inflammation, hs-CRP was measured in fasting serum and no association was found between vegetable dose and hs-CRP. Also, it was indicated that there was no relationship between total plasma carotenoids and hs-CRP.

Faraj *et al.* (2010) conducted a hypocaloric dietary intervention study to determine if there was an association of the reduction in inflammation and insulin resistance following an energy restriction diet with a decrease in serum apoB-lipoproteins in postmenopausal women who were overweight and obese (BMI $\geq$27 kg/m^2, n=56). The hypocaloric intervention aimed to reduce participants' body weight by 10% and lasted 6 months. Restricted energy requirements were calculated based on resting metabolic rate and a balanced diet (55% carbohydrate and 30% fat) were prescribed to the participants. Before and after the intervention period, women were placed on weight-stabilisation for a month. Regarding inflammatory markers, levels of hs-CRP, orosomucoid, and haptoglobin were assessed. There was a significant reduction in weight (84.0±14.3 vs 78.6±13.5 kg, $p<0.0001$), hs-CRP (3.1±2.3 vs 2.5±2.1 mg/L, $p<0.05$), orosomucoid (0.9±0.2 vs 0.8±0.2 g/L, $p< 0.01$), and haptoglobin (1.3±0.5 vs 1.1±0.4 g/L, $p< 0.01$) at the end of this intervention study. Also, it was reported that there was a strong and independent correlation between the reduction in apoB-lipoproteins and the decrease in inflammatory markers and insulin resistance in overweight/obese women when they followed a hypocaloric diet.

Feng *et al.* (2013) undertook a randomised, double-blinded, placebo-controlled trial to investigate the efficacy of histidine supplementation on insulin resistance, inflammation, oxidative stress and metabolic disorders in obese women (BMI ≥28 kg/m^2) with the metabolic syndrome. For the intervention, the women were randomly assigned to either histidine (n=50) or placebo group (n=50). The histidine group received 4 g histidine daily (two 1 g histidine tablets after breakfast and two tablets after dinner) for 12 weeks, whereas the placebo group received four tablets of placebo (just containing maize starch) daily during the same period. In terms of inflammation, biomarkers of TNF-α, IL-6, and CRP were measured at baseline and 12 weeks, with no significant differences in these variables reported between the two groups at baseline. At the end of the intervention, whilst serum histidine significantly increased, TNF-α (3.96 pg/ml; 95%CI: −5.29, −2.62) and IL-6 (2.15 pg/ml; 95% CI: −2.52, −1.78) significantly decreased between baseline and 12 weeks in the histidine supplementation group. Also, in the same group, a significant reduction in BMI (0.86 kg/m^2; 95% CI: −1.55, −0.17) and waist circumference (2.86 cm; 95% CI: −3.86, −1.86) was reported.

Ghavipour *et al.* (2013) assessed the effects of tomato juice consumption on inflammation in overweight or obese women (BMI ≥ 25 kg/m^2). The participants (n=106) were randomly allocated to the intervention or control group. The intervention group consumed 330 ml of tomato juice (including 37.0 mg lycopene) daily for 20 days, whilst the control group maintained their usual diet with water during the same period. IL-6, IL-8, TNF-α, and hs-CRP were measured to assess inflammation. Following the intervention period, serum concentration of TNF-α and IL-8 decreased significantly in the intervention

group compared with baseline and the control group. Whilst there was a significant decrease in TNF-α and IL-8 among overweight subjects in the intervention group compared with baseline, no differences in IL-8, TNF-α and hs-CRP were reported in obese subjects in the same group.

Kazemzadeh *et al.* (2014) conducted a randomised crossover clinical trial to compare the impact of white rice and brown rice on hs-CRP and cardiovascular risk factors in non-menopausal females who were overweight or obese (BMI > 25 kg/m^2). The study comprised two intervention periods of two groups, group 1 and 2. The participants in group 1 (n=20) consumed brown rice in the first period and white rice in the second period, whereas those in group 2 (n=20) consumed white rice in the first period and brown rice in the second period. Each intervention period lasted 6 weeks, with a 2-week washout phase between the intervention periods. In both intervention periods, the women were asked to follow a hypocaloric diet (200 to 500 kcal/day, 150 g brown or white rice). The results showed that the brown rice diet in comparison with white rice significantly reduced weight (1.2 kg; 95% CI = 0.60–1.96, p=0.001), waist circumference (2.38 cm; 95% CI = 0.51–4.25, p=0.014) and hs-CRP (0.98 mg/L; 95% CI = 0.23–1.73, p=0.012). In terms of lipid profiles and fasting blood glucose, there was no significant difference between the two diets.

Rosado *et al.* (2011) conducted a randomised, controlled intervention study to investigate the effect of the intake of low-fat milk and low-fat milk with added micronutrients on anthropometrics, body composition, blood glucose levels, lipids profile, CRP, and blood pressure of women (BMI ≥30 kg/m^2) following an energy-restricted diet. Participants were randomly assigned into three groups:

250 ml of low-fat milk consumed three times per day with a hypocaloric diet (500 kcal/day) (n=46), 250 ml of low-fat milk with added micronutrients (iron, magnesium, vitamin E, niacin [3.11 mg/dose], folic acid, thiamine, vitamin B-6, and vitamin B-12) consumed three times per day with a hypocaloric diet (500 kcal/day) (n=46), and an energy-restricted diet (500 kcal/day) with no intake of milk (n= 47) for 16 weeks. After the 16-week intervention, the greatest weight loss was observed in women who consumed low-fat milk with added micronutrients (5.1 kg; 95% CI: 6.2–4.1). In terms of inflammation, no differences were found between groups in level of CRP. Also, there was no effect observed on glucose level, blood lipid profile or blood pressure.

Ryan *et al.* (2014) undertook a prospective controlled study to assess the association of plasma and tissue markers of systemic and vascular inflammation with obesity and insulin resistance and compared the effects of aerobic exercise training plus weight loss versus weight loss alone on markers of inflammation in obese adult women (BMI 25–46 kg/m^2). Before the weight loss intervention, all subjects (n =77) were required to follow a therapeutic lifestyle changes diet for 6–8 weeks and they were encouraged to consume low dietary fat, high fibre and low sodium diet. Afterwards, all subjects were required to attend weekly weight loss classes which aimed to restrict calorie intake by 300 to 500 kcal/d to achieve weight loss for 6 months. At the same time, they maintained the macronutrient composition of their diet by following the therapeutic lifestyle changes diet. The women in the aerobic exercise training plus weight loss group exercised 3 times per week (45 min) for 6 months. As a biomarker of low-grade inflammation, CRP was measured before and after the interventions, showing

that CRP reduced by 29% with aerobic exercise training plus weight loss ($p < 0.001$) and 21% with weight loss alone ($p<0.05$).

2.2.3.2 Key Findings

Six trials in this review investigated the effects of hypocaloric diets either alone (Faraj *et al.*, 2010) or combined with other interventions including a nutritional supplement (Alves *et al*., 2013), higher and lower protein (Cheng *et al.*, 2013), brown rice (Kazemzadeh *et al.*, 2014), low-fat milk with added micronutrients (Rosado *et al.*, 2011), and aerobic exercise (Ryan *et al*., 2014). Four interventions examined other dietary approaches including freeze-dried strawberry powder (Basu *et al*., 2009), vegetable intake (Crane *et al*., 2011), histidine supplementation (Feng *et al*., 2013), and tomato juice (Ghavipour *et al*., 2013).

All intervention studies that included a hypocaloric approach achieved weight loss and four of those found positive effects of the interventions on biomarkers of inflammation in overweight or obese adult women (Cheng *et al*., 2013; Faraj *et al*., 2010; Kazemzadeh *et al*., 2014; Ryan *et al*., 2014). Among other dietary approaches, histidine supplementation (Feng *et al*., 2013) and tomato juice (Ghavipour *et al*., 2013) were found to be effective in improving biomarkers of inflammation. In total, six out of ten interventions had positive impacts on inflammation.

2.2.4 Discussion

The present study was designed to explore the effectiveness of interventions designed to address inflammation in overweight or obese adult women. This

review found that weight loss may be associated with a decline in some biomarkers of inflammation, such CRP, in adult women who are obese. Also, higher protein (Cheng *et al*., 2013), brown rice (Kazemzadeh *et al*., 2014), aerobic exercise (Ryan *et al*., 2014), histidine supplementation (Feng *et al*., 2013), and tomato juice (Ghavipour *et al*., 2013) were found to be effective in reducing inflammation with or without weight loss.

CRP was a common biomarker of inflammation measured in all studies included in this review and most interventions that achieved weight loss effectively reduced the concentration of CRP in women with obesity. This finding broadly supports the findings of other studies in this area linking CRP with weight loss. Selvin *et al*. (2007) conducted a systematic review to assess if weight loss was directly related to a reduction in CRP level. Analysis of data (the mean change in CRP level and the mean weight change) from baseline and post-intervention from 33 included studies showed that there was a relationship between weight loss and a decline in CRP level; for each 1 kg of weight loss, the mean change in CRP level was reported as -0.13 mg/l.

Two separate systematic reviews were also performed to clarify the effects of flaxseed interventions and plant-based diets on inflammation. In the meta-analysis by Ren et al. (2016), there was insufficient evidence that flaxseed and its derivative have a beneficial effect on reducing circulating CRP. However, it was reported that they may significantly reduce CRP in obese populations. Another meta-analysis of intervention trials to assess the effect of plant-based diets on obesity-related inflammatory biomarker profiles found that consumption of plant-based diets was associated with a reduction in the mean

concentrations of some inflammatory markers including CRP and IL-6. However, no substantial effects were revealed for TNF-α, adiponectin and leptin (Eichelmann *et al*., 2016).

Although systematic reviews that assess the effect of a variety of factors, such as weight loss (Selvin *et al*., 2007), exercise (Simpson & Singh, 2008), and consumption of dairy products (Labonte *et al*., 2013) were available, the importance and originality of this study is that it explores the effect of all kinds of interventions including hypocaloric diets, physical activity, and dietary supplementations on inflammation in only adult women who are overweight or obese.

All trials in this review assessed obesity-related inflammation, but only one study (Cheng *et al*., 2013) measured plasma hepcidin level. Cheng *et al*. (2013) measured plasma hepcidin only at baseline, surprisingly it was within normal range for overweight and obese participants (BMI ≥27.5 kg/m^2). As hepcidin-mediated iron disturbances were not proven at baseline, this marker was not measured at follow-up. Although these findings cannot be extrapolated to all people who are obese, the benefit of weight loss on iron status may not be independently related with a reduction in hepcidin circulation. Thus, further research should be undertaken to investigate the effect of weight loss on elevated hepcidin levels in obese subjects. Also, to develop a full understanding of the relationship between low-grade inflammation due to excess adiposity and hepcidin, anti-inflammatory intervention studies are needed to measure hepcidin levels in obese subjects, so as to develop targeted interventions aimed at ID in women who are obese.

A considerable amount of literature paid particular attention to the effects of plant-based diet which could be associated with a lower BMI attributable to the lower energy density of the diet on inflammation (Eichelmann *et al.*, 2016). Plant-based diets are associated with higher dietary fibre intake. A meta-analysis by Jiao *et al*. (2015), reported that dietary fibre or food naturally rich in fibre has beneficial effects on circulating CRP level in overweight/obese adults. Another potential explanation for the observed association of plant-based diet with changes in inflammatory markers might be its effect on reducing oxidative stress which is associated with saturated fat (Lüscher, 2015). Based on the relationship between oxidative stress and inflammation, Ghavipour *et al.* (2013) investigated the effects of tomato juice consumption. Higher lycopene content of tomato juice is a natural pigment, the most potent antioxidant and protects the body against oxidative stress by neutralizing free radicals (Chaiyasit *et al.*, 2005). Improving effects of dietary fibre and antioxidants on inflammation brought about an increased interest in flax-seed which has a high fibre component and antioxidant capacity (Ren *et al*., 2016). Similarly, Feng *et al*. (2012) focused on histidine because low plasma concentrations of histidine are associated with oxidative stress, inflammation and increased mortality in patients with chronic kidney disease (Watanabe *et al*., 2008).

In conclusion, these two systematic style reviews revealed that weight loss, increasing PA and reducing energy-dense foods by consuming more fibre and antioxidant rich foods, may play important roles in both improving iron status and reducing inflammation in people who are obese. However, both systematic style reviews were subject to a number of potential methodological weaknesses. The majority of the stages associated with conducting the review (selection of

primary studies, study quality assessment and data extraction) were carried out by only one researcher (HKK). Secondly, a clear data synthesis was not able to be done, due to the methodological heterogeneity of the included studies such as assessment of iron status and inflammation.

The key findings from this chapter provided additional detail about the current gaps and knowledge especially in terms of the importance of weight management, PA and reducing energy-dense foods. Very little is currently known about health consequences of a combination of obesity and ID. Also, much uncertainty still exists about the potential links between weight, iron status, dietary factors and PA. The research questions of study 1 were shaped, based on the finding from these two systematic style reviews (which are outlined in section 1.2.4.2 in chapter 1).

CHAPTER 3: A SURVEY INVESTIGATING DIET AND FATIGUE IN FEMALE SLIMMING WORLD MEMBERS

Obesity and iron deficiency appear to be linked with significant lower mean serum iron concentration in overweight compared with normal-weight individuals (Cepeda-Lopez *et al*., 2010). Iron deficiency anaemia is generally associated with reduced 'get-up-and-go' and a higher level of fatigue (Acaster *et al*., 2015) (see section 1.1.5 in chapter 1). This chapter presents both methods and results of study 1 which aimed to identify any links between BMI, fatigue level, dietary patterns and dietary factors affecting the bioavailability of iron and physical activity levels using an online survey. The research questions previously listed (see section 1.2.4.2 in chapter 1) were investigated with the following objectives:

I. To determine BMI and level of fatigue in a population of SW members before they started Food Optimising (T0), whilst they are still a new member (around 4 weeks after joining) (T1) and 12 weeks later (T2) and identify weight and fatigue changes over these time periods.
II. To determine physical activity levels in women at T0, T1 and T2 and identify changes over these time periods.
III. To explore associations between fatigue level, physical activity level and weight change in women at T0, T1 and T2.
IV. To identify the most frequently consumed food groupings in women at T0, T1 and T2.
V. To determine dietary habits in women at T0, T1 and T2

VI. To determine iron-related dietary patterns of women who attended SW at T0 and T1.

VII. To explore associations between BMI, level of fatigue and the dietary patterns at T0 and T1.

3.1 METHOD

3.1.1 Study design

A pragmatic indicator of ID was required because obtaining information regarding diagnostic biomarkers of ID was not possible. A validated questionnaire to assess level of fatigue, one of the main symptoms of ID was sourced. Since the research question was multifactorial, a combination of validated questionnaires were selected to measure food frequency, dietary factors affecting the bioavailability of iron and physical activity levels.

Between March and May 2016, the most appropriate validated tools available to as accurately as possible report on fatigue level, food frequency, dietary patterns and dietary factors affecting the bioavailability of iron, and physical activity level was established.

3.1.2 Ethical considerations

Ethical approval was obtained from University of Nottingham School of Biosciences research ethics committee (SBREC150132A) on 18/07/2016 (see appendix M).

Information about the study including aim of the study, benefits and risks of taking part, the kinds of questions to be asked, as well as confidentially of data collection were explained on the first page of the online survey. On the second

page, there was an online consent form. Women who agreed to participate in the study accessed the questionnaires after confirming their consent online.

3.1.3 Sampling

The study population was women of child-bearing age (19-49 years), not pregnant and with a starting BMI ≥25 kg/m^2.

In order to access a representative population of overweight and obese people who wish to lose weight, Slimming World was chosen as the setting for participant recruitment. Also, it was a suitable organisation to facilitate their members to complete an online survey. It has been suggested that commercial slimming organisations provides an effective and resource friendly environment for recruiting volunteers who are either overweight or obese (Randell *et al.* 2015). Female members who had recently joined SW (less than 8 weeks) were invited to participate in the study.

3.1.4 Questionnaire development

The survey consisted of four sections: socio-demographic characteristics and general health and well-being questionnaire; fatigue questionnaire; food frequency and dietary habits questionnaire; and physical activity questionnaire (see appendix N).

3.1.4.1 Socio-demographic characteristics and general health and well-being questionnaire

Self-reported questions in this section included about participants' date of birth (ages were calculated based on survey opening date), height in feet and inches or centimetres, weight in stone and pounds or kilograms, weight measurement

method, smoking status, number of pregnancies and general women's health and well-being. Concerning ID, women were asked if they had ever suffered from low iron stores, ID or IDA and treated for ID or IDA.

3.1.4.2 Questionnaire selection to measure level of fatigue

An instrument that could be used to obtain internationally comparable data was required because it was planned to use same survey questions in studies in the UK and Turkey. Also, since collecting data relating to before joining Slimming World (BSW) (T0) and current situation (T1) at the same time (in one survey) was planned, recall periods for items included in questionnaire should be validated as short as possible to allow participants make a comparison between T0 and T1.

There are a number of instruments available for measuring fatigue but none of them was specifically designed to investigate fatigue in healthy obese population. Hence, the instruments that allow to assess fatigue in general population were considered. Chalder Fatigue questionnaire and Short Form 36 (SF-36) were considered but not selected because of their long recall periods for items (4 weeks).

The Multidimensional Assessment of Fatigue (MAF) (Belza *et al.,* 1995) was selected. A modification of the Piper Fatigue Scale demonstrated face and content validity (Piper *et al.,* 1998) and aimed to evaluate self-reported fatigue in patients with rheumatoid arthritis (Belza *et al*., 1995). However, the MAF subsequently used to assess fatigue in diverse range of disease. Turan *et al*. (2007) tested the MAF against SF-36 in ankylosing spondylitis patients and found that there was a moderate correlation of MAF was with activity pain

(r=0.39), Hb (r= -0.38) and all dimensions of SF-36 except for social function and emotional role. Similarly, Yildirim & Ergin (2013) tested the MAF against SF-36 and determined the reliability and validity of the Turkish language version of the MAF in chronic musculoskeletal physical therapy patients (n=69). Findings from the cronbach's alpha and the correlations between the total scores of the Turkish language version of the MAF scale and the subscale scores of SF-36 indicated that the Turkish language version of the MAF was a valid and reliable scale to measure fatigue in chronic musculoskeletal physical therapy patients.

This validated questionnaire measures self-reported fatigue with; degree and severity (items 1,2), the amount of distress it causes (item 3), the degree to which fatigue interferes with activities of daily living including household chores, cooking, bathing, dressing, working, socializing, sexual activity, leisure/recreational, shopping, and walking (items 4-14), and timing (its frequency and changes in its degree) (items 15,16). Respondents were asked to express their fatigue patterns for the past week. Numerous aspects of fatigue are included in the questionnaire in order to produce a global score. Because only the first 15 items are used to calculate the global fatigue index (GFI), the 16th item which asks if it changed over the past week was not included in both the first and second survey.

The MAPI Research Trust was contacted to ask permission and obtain the questionnaire in two different languages as English (UK) and Turkish. User agreement was signed and permission to use the questionnaire was obtained (April 2016).

3.1.4.3 Questionnaire selection to measure food frequency and dietary habits

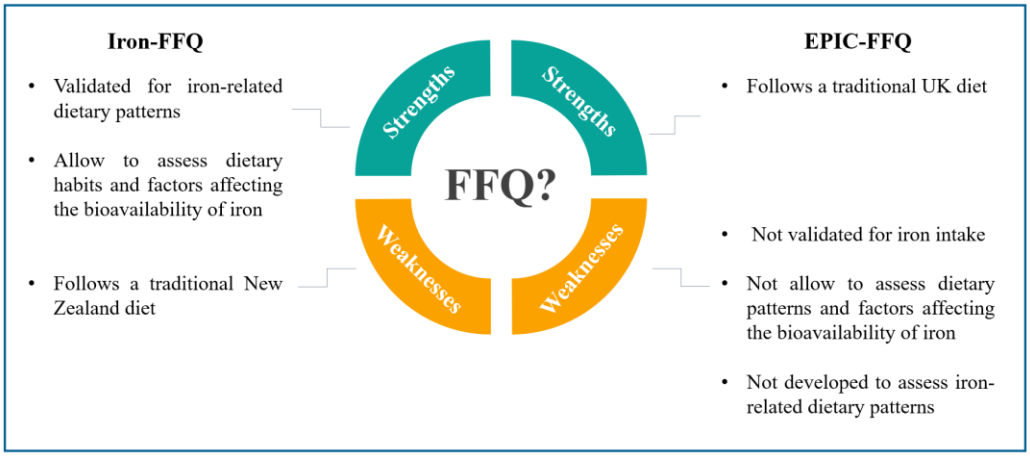
The strengths and weakness of two different food frequency questionnaire (FFQ), the EPIC-Norfolk Food Frequency Questionnaire (Bingham *et al*., 2001) and an iron FFQ (FeFFQ) developed by Beck *et al*., (2013) were considered (see figure 3.1). It was decided to use FeFFQ (Beck *et al*., 2013), because of the following reasons;

The questionnaire was divided in two parts. The first part was a qualitative FFQ that contained 144 food groupings (each corresponding to a single question) divided into the following 15 food categories: meat, fish and chicken; prepared meat; seafood; fruit; vegetables; legumes; breakfast cereals; breads, cakes, biscuits and crackers; grains; dairy products; eggs; nuts; miscellaneous foods and snacks, alcohol; and non-alcoholic beverages. The purpose of this part of the questionnaire was to get an idea on the overall consumption pattern of the participants. In order to define certain clusters of food consumption, participants were asked how often they consumed items from each food grouping. Suggested portion sizes were included for each food groupings. Options for frequency of intake included never or less than once per month, one to three times per month, once per week, two to three times per week, four to six times per week, once per day, two to three times per day, and four plus times per day. The second part of the iron-questionnaire focused on food habits/behaviour. This part enabled the researchers to gain more information on how the participants consume the different food items that were included in the first part.

The FeFFQ was specifically designed to identify iron-related dietary patterns and validated in New Zealand, an OECD country with one of the highest prevalence rates of obesity (see section 1.16.1 in Chapter 1). In order to evaluate its relative validity and reproducibility, young women aged 18 to 44 years (n=115) completed the FeFFQ at baseline and 1 month later. Between these assessments, they were asked to complete a 4-day weighed diet record. Identified dietary patterns from the two FeFFQs and the 4-day weighed diet record indicated that the FeFFQ was a reproducible and relatively valid method for determining dietary patterns, and could be used to investigate the correlation between dietary patterns and iron status (Beck *et al*., 2012).

The lead author (Beck) for the validation of the iron questionnaire was contacted and permission to use the questionnaire was obtained (March 2016).

Figure 3.1 Pros and cons table for FFQs



3.1.4.4 Physical activity questionnaire

Similar to fatigue, food frequency and dietary habits questionnaire, an instrument that could be used to obtain internationally comparable data was required. Also, a questionnaire that allows a self-administrated method was required. Since the international physical activity questionnaire (IPAQ) met these requirements, it was selected. The short form, with seven questions, that recorded the activity at four levels (vigorous-intensity activity, moderate-intensity, walking, and sitting) was used to prevent the overall questionnaire being excessively long.

The IPAQ was publicly available with open access. No permission was required to use it.

3.1.5 Data collection

The survey was uploaded using the Bristol Online Survey tool after discussions with the SW research team to ensure that they were happy with the study protocol and some questions were modified to avoid weight stigmatisation. The first survey was designed to collect data of participants before joining SW (T0) and their current habits (NOW) (T1) at the same time. The first survey included 220 questions and the second survey included 192 questions in total. Estimated time required to complete the surveys were 45 and 30 minutes respectively for the first and second surveys. The link for the online survey was available on the SW member's website for two weeks as of the beginning of the data collection (July 2016). 12 weeks after the first survey (T2), the same participants were invited to complete the second survey via e-mail. Some questions from the section of socio-demographic characteristics and general health and well-being

questionnaire on the first survey such as date of birth, height, smoking status and description of eating pattern were not asked again on the second survey. The fatigue questionnaire (MAF), FeFFQ, and short form physical activity questionnaire (IPAQ) were the same as the first survey.

3.1.6 Data Analysis

3.1.6.1 Measures

3.1.6.1.1 Weight, height and body mass index

In the first survey, height was self-reported as feet and inches or centimeters. In both first and second survey, body weight was self-reported as stones and pounds or in kilograms. Microsoft Excel (2016) was used to convert the weights which were in stones and pounds into kilograms and the heights which were in feet and inches into metres. Body mass index was calculated as weight (kg)/ height (m) squared for the three time periods.

3.1.6.1.2 Global fatigue index (GFI)

In order to calculate GFI, 15 items from the MAF questionnaire were used. Numerical rating scale was 1-10 for item 1-14 and responses were categorical (from every day to hardly any days) for item 15.

In accordance with the methodology described by Neuberger (2003), each score for item 15 was multiplied by 2.5 and converted to a 0-10 scale. Items 1,2, and 3, average of items 4-14, and newly scored item 15 were summed. If respondents reported that they "do not do any activity for reasons other than fatigue", a zero score to items was assigned. If respondents reported "no fatigue" on item 1, a

zero to items 2-15 was assigned. For GFI, higher score represented higher level of fatigue and the score range was 1-50 (1= no fatigue, 50= severe fatigue).

3.1.6.1.3 Cut-off score for GFI

A cut-off score was used to discriminate between cases and non-cases for presence of fatigue. Although GFI has been used to define fatigue level in a diverse range of disease states and healthy populations, a specified cut-off score for women of child-bearing age did not exist. The initial testing of the MAF was carried out by Belza *et al.* (1995) in patients with rheumatoid arthritis (n=51) and healthy controls (n=46). Whilst mean GFI in rheumatoid arthritis patients was 29.2, 28.1, and 26.1 at 3 time periods 6 – 8 weeks apart, it was 17, 16.5, and 15.8 at the 3 time periods in healthy controls.

Cantor *et al.* (2008) used the same fatigue questionnaire (MAF) as Belza *et al.* (1995). In this study, the cut-off score was defined as 21 to examine the extent of clinically significant fatigue in the sample (individuals with mild to severe traumatic brain injury [n=223] and non-injured controls [n=85]). This score was chosen because it is half a standard deviation (SD=11) below the mean score for individuals with rheumatoid arthritis in the study by Belza *et al.* (1995) and half a SD above the mean score for healthy controls. Using this cut-off score, it was shown that whilst 75% of individuals with traumatic brain injury had significant fatigue, only 40% of those in the control group had significant fatigue (chi square (df=1, n=308) =34.05, $p<0.001$).

In another study (Englander *et al*., 2010) employing the same questionnaire, a score of 27 and above was used as the cut-off for the presence of fatigue in the convenience sample of 119 individuals at least 1-year post-traumatic brain

injury. Using this cut-off score, it was found that more than half of the participants (53%) presented with fatigue. Similarly, Anandacoomarasamy *et al*. (2009) used the MAF questionnaire and GFI in their study to determine health-related quality of life and fatigue measures in participants with obesity and to compare scores with age- and gender-matched population norms. A cut-off score for GFI was not defined in this study. The results showed that mean GFI was 24.9 (±12.6). Because age- and gender-matched population norms were not yet available for the GFI, this result was compared with a control group in the study by Belza *et al.* (1995). Mean fatigue score in this study was significantly higher than scores in the control group (GFI = 17, $p<0.0001$).

In a randomised controlled study by Thorp *et al.* (2014), fatigue was assessed using a modified version of the Individual Strength Questionnaire and the MAF Scale in overweight/obese office workers (n=23; BMI; 29.6±4 kg/m^2). Because a score of ≥30 was one SD above the mean score of healthy controls in the study by Belza *et al.* (1995) and a predefined score was not available, this score was selected for clinically significant fatigue. There was no statistically significant difference between SIT condition (seated work posture) and STAND-SIT condition (interchanging between a standing and seated work posture every 30 min using an electric, height-adjustable workstation) in terms of GFI (15.8 vs 13.0, p=0.47).

Grady *et al*. (1998) also calculated GFI to assess fatigue level in a cohort of men infected with HIV (n=50). Participants were randomised to receive or not to receive investigational interleukin-2 and fatigue was measured at baseline, at the end of 5 days of interleukin-2 therapy, 1 week later at home, and 1 month later

for three consecutive cycles of interleukin-2 therapy. It was suggested that mean GFI at baseline increased after initiation of interleukin-2 treatment (from 11.5 to 27.7).

In the present study, a cut-off score was required to define presence of fatigue in women of child-bearing age (19-49 years), not pregnant and with a starting BMI ≥25kg/m^2. All the studies reviewed so far suffer from the fact that a defined cut-off score for GFI for all population groups does not exist. Hence, in our study, a cut-off score was determined based on the studies with the most similar study populations (Anandacoomarasamy *et al.*, 2009; Thorp *et al.*, 2014). Belza *et al.* (1995) was also considered given their work was instrumental in developing and validating the MAF. These studies enabled us to identify a cut-off score between 25 and 30, with a score of 28 determined for our GFI.

3.1.6.1.4 Associations between body weight, fatigue and levels of physical activity

All analyses were conducted using the IBM Statistical Package for the Social Sciences (SPSS) version 24.0 for Windows (IBM Corp., Armonk, NY) with statistical significance assumed at $p \leq 0.05$. A one-way repeated measures ANOVA was conducted to identify changes in BMI, fatigue level and physical activity level over the three time periods (T0, T1 and T2) for the 74 participants with data available for all three time-points.

The association between weight status, fatigue level and physical activity level was investigated by Multiple Regression models. The first model was built to examine the relationship between percentage weight change and various potential predictors including baseline BMI, fatigue level (T0 and T2), and

physical activity level (T0 and T2). A hierarchical multiple regression was built as the second model to understand if baseline BMI and decrease in fatigue level predict weight loss, after controlling for the influence of physical activity at T0, T1 and T2. For the third model, the change in fatigue level was identified as a continuous dependent variable in order to predict how weight loss and physical activity levels influence this change.

3.1.6.1.5 Dietary patterns

Responses from the FeFFQ were converted into eight frequencies of intake per week for each food grouping for each participant, ranging from zero to 28 times eaten per week. The average weekly consumption of each food grouping was determined, and the 35 most frequently consumed food groupings (as number of times consumed per week) were identified for use in factor extraction.

Factor extraction is a technique which allows the researcher to determine the smallest number of factors that can be used to best represent the interrelationships among the set of variables. Principal components analysis (PCA) is the most commonly used approach for factor extraction (Pallant, 2016). The number of factors that are considered best describes the underlying relationship among the variables should be determined according to balancing two conflicting needs: the need to find a simple solution with as few factors as possible; and the need to explain as much of the variance in the original data set as possible (ibid).

First, the Kaiser-Meyer-Olkin Measure of Sampling Adequacy (KMO) value was checked to verify if the data set was suitable for factor analysis. To

determine how many components (factors) to 'extract', three different techniques (Kaiser's criterion, scree test and parallel analysis) were interpreted.

Kaiser's criterion

This is also known as the eigenvalue rule and according to this, only components that have an eigenvalue of 1 or more should be considered. Each eigenvalue represents the amount of variance that has been captured by one component (Wuensch, 2012)

Scree test

The scree test involves plotting each of the eigenvalues of the factors and inspecting the plot to find a point at which the shape of the curve changes direction and becomes horizontal (Pallant, 2016). Catell (1966) suggested that all factors above the elbow or break in the plot must be retained because these factors contribute the most to the explanation of the variance in the data set.

Parallel analysis

Horn's parallel analysis (Horn, 1965) is an additional technique to identify the correct number of factors to retain. In order to carry out parallel analysis, another statistical program (developed by Watkins, 2000) that is called Monte Carlo PCA for Parallel Analysis was used. The programme generated 100 sets of random data of the same size as the real data file and calculated the average eigenvalues for these 100 randomly generated samples. The size of the eigenvalues was compared with those obtained from a randomly generated data set of the same size. Only those eigenvalues that exceed the corresponding values from the random data set were retained.

3.1.6.1.6 Physical activity

Continuous indicators obtained from the IPAQ are presented as total physical activity Metabolic Equivalent of Task (MET)-minutes/week. An average MET value for each type of activity (walking, moderate-intensity and vigorous intensity was derived from the compendium (Ainsworth *et al*., 2000). The following values were used to analyse IPAQ data:

Walking MET-minutes/week = 3.3 x walking minutes x walking days

Moderate MET-minutes/week = 4.0 x moderate-intensity activity minutes x moderate days

Vigorous MET-minutes/week = 8.0 x vigorous-intensity activity minutes x vigorous-intensity days

Total physical activity MET-minutes/week = sum of Walking + Moderate + Vigorous MET-minutes/week scores (IPAQ Research Committee, 2005).

3.1.6.1.7 Dietary habits

Data from the dietary habits questionnaire was coded as "yes" or "no". Percentages of "yes" for each item, in the dietary habits questionnaire, were listed at T0, T1 and T2. In order to compare findings between T0 and T1, and T1 and T2, McNemar's test (a 2x2 cross classification of paired (or matched) responses to a dichotomous item (Adedokun & Burgess, 2012)) was used. The test showed decision as "reject/retain the null hypothesis" with $p \leq .05$ indicating statistically significant differences between T0 and T1 or T1 and T2. The null

hypothesis was “the distribution of different values across T0 and T1 or T1 and T2 are equally likely”.

3.2 RESULTS

Subject recruitment was carried out from July to November 2016 (see figure 1.12 in Chapter 1).

Eight hundred and twenty-five SW members completed the first survey with 282 excluded from the analysis because; 257 women were ≥50 years old, 3 women were ≤18 years old and 22 women joined SW ≥8 weeks ago. 543 participants were eligible for data analysis at T0 and T1.

One hundred and sixty-six SW members completed the second survey and 92 of those were excluded from the analysis because either their e-mail addresses or their SW membership number did not match with responses from the first survey (date of birth was not included in the second survey which could have helped match up records). Therefore, data of 74 women was available for analysis over the three time points (T0, T1 and T2).

3.2.1 Sample description

The characteristics of the 543 participants at T1 and 74 participants at T2 are presented in Table 3.1. Since data of participants before joining Slimming World (T0) and their current habits (T1) were collected at the same time, their mean number of weeks as a SW member (2.5 weeks) represented the mean time intervals between T0 and T1. The minimum time interval between T1 and T2 was 12 weeks and the maximum was 22 weeks.

The majority of women (mean age 37 years) were non-smokers, were eating a variety of all foods including animal products, were healthy with no/little chronic disease and with no/little ID, were pre-menopausal and were having

moderate period losses with a period duration of approximately five days. The majority had experienced one full-term pregnancy.

In terms of iron status, whilst approximately half of women (52.1%) reported that they had never suffered from low iron stores, ID or IDA at T1, three-quarters of those (75.3%) reported that they had never suffered from low iron stores, ID or IDA in the last three months on the second survey (at T2). Regarding treatment for ID or IDA, majority of women indicated that they had never received a treatment at T1 (59.8%) and in the last three months (94.3% at T2).

Table 3.1 Characteristics of the study population in Study 1

	T1 (n=543)	T2 (n=74)
Mean age (years)	36.7 (±8.4) [19-49] (n=503)	-
Mean number of weeks as a SW member	2.5 (±1.4) [1-7] (n=500)	-
Weight measurement method (%)		
Measured on scales at SW group	89.0 (n=453)	83.8 (n=62)
Bathroom scales	8.4 (n=43)	13.5 (n=10)
Smoking status (%)		
Yes	10.2 (n=52)	-
No	68.9 (n=352)	-
An ex-smoker	20.5 (n=105)	-
Eating pattern (%)		
Eat a variety of all foods, including animal products	87.1 (n=446)	-
Eat eggs, dairy, fish and chicken but avoid other meats	2.3 (n=12)	-
Eat eggs but avoid dairy products, all meats and fish (ovo-vegetarian)	0.8 (n=4)	-
Eat dairy products but avoid eggs, all meats and fish (lacto-vegetarian)	0.0 (n=0)	-
Eat eggs and dairy but avoid all meats and fish (ovo-lacto vegetarian)	2.3 (n=12)	-
Eat no animal products including eggs, dairy, and honey (vegan)	1.6 (n=8)	-
Eat fish or other sea food but avoid all other meats (pescatarian)	2.3 (n=12)	-
Other	3.3 (n=17)	-
Acute or chronic illness status (%)		
Yes	13.9 (n=71)	8.1 (n=6)
No	85.3 (n=436)	89.2 (n=66)
Low iron stores, ID or IDA status (%)		
Yes	37.8 (n=193)	9.6 (n=7)
No	52.1 (n=266)	75.3 (n=55)
Receiving a treatment for ID or IDA (%)		
Yes	36.9 (n=188)	5.4 (n=4)
No	59.8 (n=305)	94.6 (n=70)
Number of full-term pregnancies	1.26 (±1.3) [0-7] (n=500)	-
Post-menopausal status (%)		
Yes	6.5 (n=33)	-
No	92.0 (n=470)	-
Prefer not to answer	1.6 (n=8)	-
Mean period duration (day)	4.63 (±2.3) [0-14] (n=453)	-
Volume of period (%)		
Light	20.4 (n=96)	-
Moderate	35.1 (n=165)	-
Heavy	30.0 (n=141)	-
Very heavy	14.5 (n=68)	-

3.2.2 Changes in BMI, fatigue and PA levels over the three time points

A one-way repeated measures ANOVA was conducted to compare BMI, GFI and PA at T0, T1 and T2. The analysis indicated that there were significant differences ($p<0.001$) in BMI over the three time points (T0, T1 and T2). This results suggest a significant effect for time, Wilks' Lambda = 0.26, $F(2,68)=95.30$, $p<0.001$, multivariate partial eta squared = 0.74. Similarly, there were significant differences ($p<0.001$) over the three time points (T0, T1 and T2) in terms of fatigue level (Wilks' Lambda = 0.46, $F(2,72)=42.19$, $p<0.001$, multivariate partial eta squared = .54) and physical activity level (Wilks' Lambda = 0.65, $F(2,72)=19.49$, $p<0.001$, multivariate partial eta squared = 0.35). The means and standard deviations (SDs) are presented in Table 3.2.

Table 3.2 Descriptive statistics for BMI, GFI and PA for T0, T1 and T2

	T0		T1		T2	p value*
	n=543	n=74	n=543	n=74	n=74	
Mean BMI (kg/m^2)	35.0 (±7.1) [22.5-60.2]	33.7 (±6.6) [23.0-60.2]	34.1 (±6.9) [21.9-69.0]	33.0 (±7.6) [21.9-69.0]	31.0 (±7.2) [21.8-60.3]	<0.001
Mean GFI	29.2 (±12.8) [1-50]	29.4 (±12.1) [1-50]	21.8 (±11.8) [1-50]	20.8 (±11.5) [1-50]	18.1 (±10.7) [1-43.9]	<0.001
Mean total PA (MET-minutes/week)	1335.1 (±2506.9)	1369.3 (±1968.8)	3000.2 (±3762.4)	3312.4 (±4649.5)	3310.9 (±3943.1)	<0.001

*Statistical significance was determined for over the three time points (T0-T1-T2, n=74)

Note: The minimum time interval between T1 and T2 was 12 weeks and the maximum was 22 weeks.

3.3.3 Associations between body weight, fatigue and levels of physical activity

The first model that examines the association of weight change with BMI, GFI and PA indicated that mean PA at T2 (MET- minutes/week) and baseline BMI was significantly and positively correlated with percentage weight loss ($r= 0.24$, $p<0.05$, $r= 0.19$, $p=0.05$). This model explained 21.9% of the variance in weight loss. GFI at T2 and PA at T2 made the strongest unique contribution to weight loss respectively (beta= -0.331, $p<0.05$; beta= -0.31, $p<0.05$) (Table 3.3).

Secondly, hierarchical multiple regression was used to assess the ability of two control measures (baseline BMI and fatigue level change as percentage over the three time points) to predict weight loss, after controlling for the influence of physical activity at T0, T1 and T2. Level of PA at T0, T1 and T2 were entered at Step 1, explaining 9.5% of the variance in weight loss. After entry of baseline BMI and fatigue level change at Step 2 the total variance of explained by the model whole was 23.5%, $F(5,66)=4.04$, $p <0.005$. The two control measures explained an additional 14% of the variance in weight loss, after controlling physical activity, R squared change = 0.139, F change $(2,66) = 6.01$, $p <0.005$. In the final model, three control measures were statistically significant, with level of physical activity at T2 recording a higher beta value (beta=0.54, $p<0.005$), baseline BMI (beta=0.24, $p<0.005$), and decrease in level of fatigue (beta= 0.31, $p<0.005$) (Table 3.3).

The third model was built to understand the association of decrease in GFI with BMI and PA. The model showed that there was a significant positive correlation between weight loss and decrease in fatigue level ($r=0.27$, $p<0.05$) (Figure 3.1).

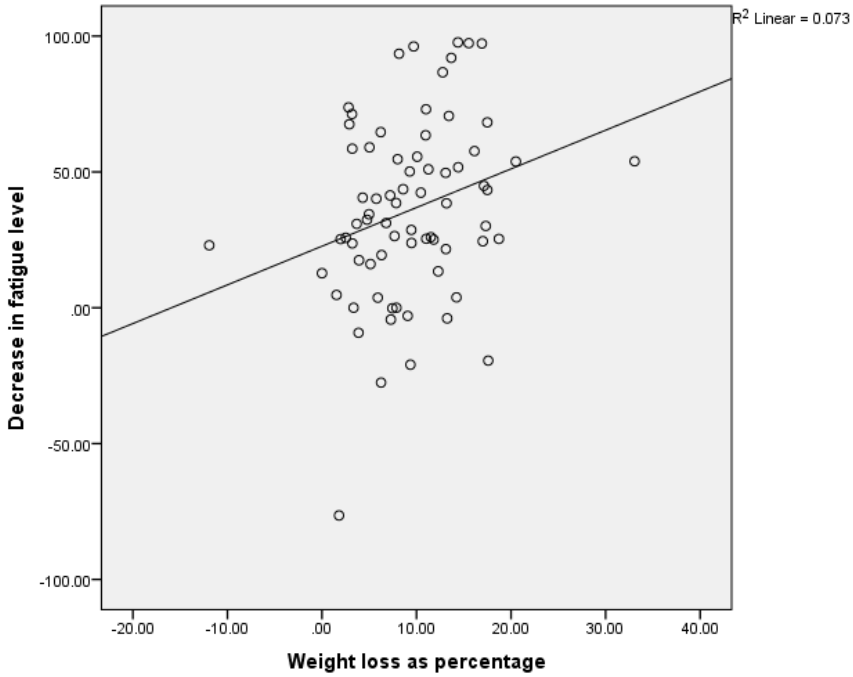
This model explained 15.3% of the variance in decrease in fatigue level and weight loss made the strongest unique contribution to explaining fatigue level change (beta= 1.6, $p<0.05$) (Table 3.3). Regarding physical activity, a significant correlation was not found between decrease in fatigue level and physical activity at any time point.

Table 3.3 Multiple regression models of weight change and fatigue level change (n=74).

Model 1							
Dependent variable	**R**	**R^2**	**Adjusted R^2**	**Independent variables**	**r**	**β**	**p value (r, β)**
Weight loss	.468	.219	.162	BMI at T0	.19	.26	0.05, 0.02
				GFI at T0	.16	.25	0.09, 0.05
				GFI at T2	-.08	-.33	0.24, 0.01
				PA at T0	-.13	-.21	0.13, 0.06
				PA at T2	.24	.31	0.02, 0.006
Model 2							
Dependent variable	**R**	**R^2**	**Adjusted R^2**	**Independent variables**	**r**	**B***	**p value (r, β)**
Weight loss	.484*	.235*	.177*	PA at T0	-.13	-.01	0.13, 0.96
				PA at T1	.08	-.35	0.25, 0.24
				PA at T2	.24	.54	0.02, 0.02
				BMI at T0	.19	.24	0.05, 0.03
				Decrease in GFI	.27	.31	0.01, 0.006
Model 3							
Dependent variable	**R**	**R^2**	**Adjusted R^2**	**Independent variables**	**r**	**β**	**p value (r, β)**
Decrease in GFI	.391	.153	.067	Weight loss	.26	1.61	0.02, 0.04
				PA at T0	.07	.05	0.28, 0.76
				PA at T1	.09	.11	0.23, 0.53
				PA at T2	-.01	-.11	0.47, 0.43
				BMI at T0	-.07	-3.50	0.30, 0.09
				BMI at T2	-.16	3.32	0.10, 0.10

*Values of step 2, after entry of BMI and fatigue level change

Figure 3.2 Scatter plot for the correlation between decrease in fatigue level and weight loss



3.3.4 Dietary patterns

3.3.4.1 Determination of dietary patterns from the iron-FFQ for before joining SW

The 35 most frequently consumed food groupings were identified for use in the PCA. The 30 most frequently consumed food groupings were identified in a study by Beck *et al*. (2013), however for the current study, because "beef" which is an important source of haem-iron was the 34th most frequently consumed food grouping at T0 (table 3.4), the 35 most frequently consumed food groupings were identified for use in the PCA.

Table 3.4 The most frequently consumed food groupings at T0

	Food groupings	Mean*	Median	SD	N
1	Milk (cow's milk) (all varieties) added to drinks	15.26	17.5	11.80	530
2	Coffee (all varieties)	10.30	7	10.88	536
3	Butter or margarine	8.56	7	7.25	535
4	Cordials (including syrups, powders)	6.24	2.5	8.47	531
5	Milk chocolate	6.03	2.5	6.87	537
6	White bread and rolls	5.86	5	6.03	536
7	Potato crisps	5.62	5	6.13	538
8	Sugar (all varieties)	5.59	0.5	8.90	533
9	Biscuits, chocolate or chocolate covered	5.36	2.5	6.86	533
10	Cooking oil (all varieties)	5.17	5	4.80	533
11	Biscuits, plain sweet	4.96	2.5	7.10	535
12	Hard cheese	4.23	2.5	4.60	533
13	Potato	3.88	2.5	2.90	540
14	Black tea	3.77	0	8.09	537
15	Milk (cow's milk) (all varieties) added to food	3.29	1	4.63	533
16	Onions (all varieties), leeks, celery	3.15	2.5	2.87	539
17	Tomatoes (all varieties)	2.84	2.5	3.27	535
18	Chicken, turkey or duck	2.73	2.5	2.12	522
19	Wholegrain bread and rolls	2.71	1	4.08	535
20	Herbal tea, fruit tea	2.50	0	6.12	536
21	Cakes	2.50	1	3.63	537
22	Yoghurt	2.37	1	3.75	535
23	Ham, bacon	2.23	1	2.37	526
24	Bananas, green bananas	2.15	1	2.78	539
25	Eggs	2.15	1	2.74	536
26	Carrots	2.00	1	2.28	536
27	Apples	1.87	1	2.99	536
28	Peppers (all varieties)	1.85	1	2.16	538
29	Milk (cow's milk) as a drink	1.82	0	4.45	534
30	Lettuce	1.81	1	2.25	536
31	Mushrooms	1.73	1	2.07	536
32	Berries (e.g. strawberries, blackberries, cherries, blueberries, boysenberries, loganberries)	1.72	0.5	3.12	539
33	Citrus fruits	1.70	0.5	3.09	536
34	Beef	1.52	1	1.97	518
35	Crackers	1.52	0.5	3.21	537

*The average weekly consumption of each food groupings based on the pre-determined portion size.

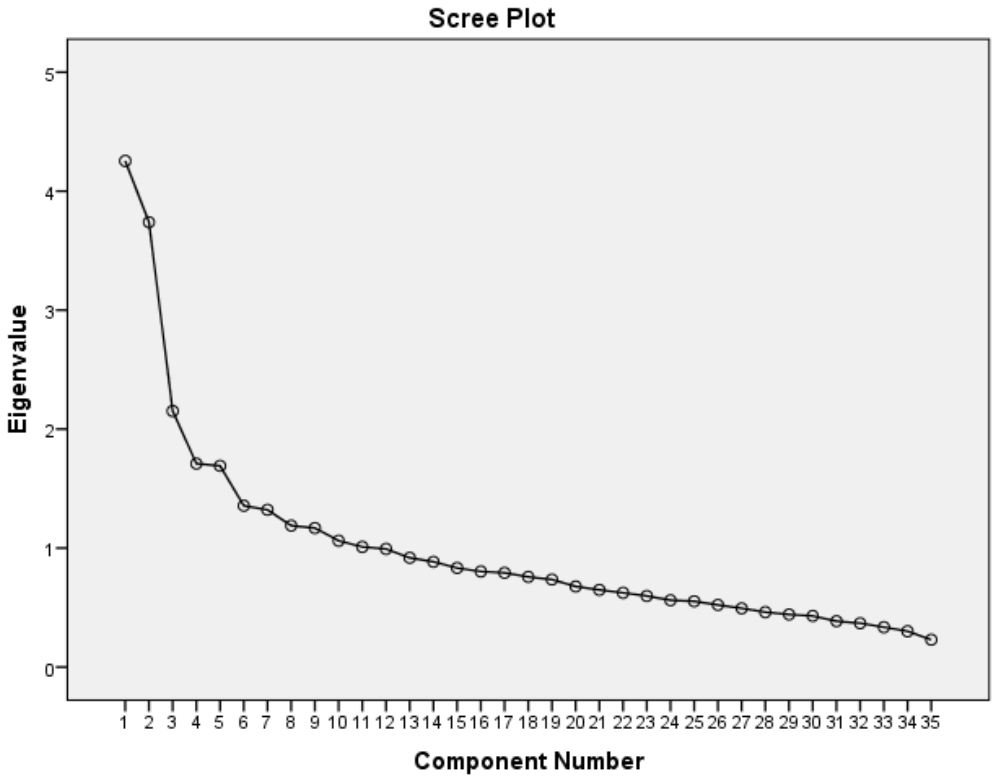
N, number of participants

Analysis of KMO indicated that the data set was suitable for factor analysis. The KMO was 0.747 (>0.6 acceptable) (Kaiser, 1970, 1974), and Bartlett's Test of Sphericity (Barlett, 1954) reached statistical significance ($p<0.001$), supporting the factorability of the correlation matrix.

3.3.4.1.1 Identifying the optimal number of factors

In order to identify the number of components to extract, the Total Variance Explained table was checked. Only the first eleven components recorded eigenvalues above 1. These eleven components explain a total of 59.00% of the variance (see appendix O). Additionally, the scree test was used to determine optimal number of factors. It was showed that there was quite a clear break between the fifth and sixth components so first five components captured much more of the variance than the remaining components (see figure 3.2).

Figure 3.3 Output from scree test at T0



Horn's parallel analysis was also used to identify the correct number of components to retain. Monte Carlo PCA for parallel analysis generated random eigenvalues from the calculated mean eigenvalues (see Parallel analysis in section 3.1.6.1.5 in Chapter 3) for 100 sets of random data for 35 variables and 543 cases (size of the real data file) (see appendix P). The first eigenvalue obtained in SPSS was systematically compared with the corresponding first value from the random results generated by parallel analysis. If the value was larger than the criterion value from parallel analysis, this factor was retained; if it was less, it was rejected. The results of parallel analysis suggested that only seven factors were retained for further investigation (table 3.5).

Table 3.5 Comparison of eigenvalues from PCA and criterion values from parallel analysis

Component number	Actual eigenvalue from PCA	Criterion value from parallel analysis	Decision
1	4.254	1.5169	Accept
2	3.739	1.4512	Accept
3	2.152	1.4042	Accept
4	1.710	1.3590	Accept
5	1.691	1.3227	Accept
6	1.356	1.2861	Accept
7	1.321	1.2558	Accept
8	1.189	1.2291	Reject
9	1.169	1.2016	Reject
10	1.062	1.1733	Reject
11	1.010	1.1473	Reject

Before making a final decision concerning the number of factors, the Component Matrix table (see appendix Q) was interpreted. The table indicated the unrotated loadings of each of the items on the 11 components which was with eigenvalues above 1 (Kaiser Criterion). The table showed that most of the items loaded quite strongly (above .4) on the first five components. Very few items loaded on Components 6, 7, 8, 9, 10 and 11. Using the default options in SPSS, an eleven-factor solution was obtained but a five-factor solution was likely to be more appropriate. Therefore, all steps were repeated for a five-factor solution.

3.3.4.1.2 Labelling the dietary patterns

According to the new Total Variance Explained table, the five-component solution explained a total of 38.7% of the variance, with Component 1 contributing 12.2%, Component 2 contributing 10.7%, Component 3 contributing 6.1%, Component 4 contributing 4.9% and Component 5 contributing 4.8%.

In order to make an easier interpretation, rotation (defined as an important and necessary step in the PCA because the factors generated by the initial unrotated solution tend to be arbitrary uninterpretable linear combinations of the true latent variables (Dien, 2010)) was applied and the factor structure were simplified. The 35 most frequently consumed food groupings were listed with the factor loadings for each dietary pattern (only factor loadings of 0.3 or higher listed) in the Rotated Component Matrix table (see appendix R) and five dietary patterns were labelled as follows:

1. **Carbohydrate (CHO) and high fat:** Milk chocolate; biscuits, chocolate or chocolate covered; potato crisps; biscuits, plain sweet; cakes; white bread and rolls; butter or margarine; potato; hard cheese; crackers; cooking oil (all varieties)

2. **Savoury staples:** Potato; onions (all varieties), leeks, celery; peppers (all varieties); lettuce; mushrooms; carrots; cooking oil (all varieties); herbal tea, fruit tea; egg s

3. **Healthy snacks:** Biscuits, plain sweet; crackers; herbal tea, fruit tea; bananas, green bananas; strawberries, blackberries, cherries, blueberries,

boysenberries, loganberries, cranberries, gooseberries, raspberries; citrus fruits; apples; yoghurt; eggs; milk (cow's milk) (all varieties) added to food

4. **Meat and eggs:** Beef; chicken, turkey or duck; ham, bacon; eggs; cordials (including syrups, powders)

5. **Milk and coffee:** Butter or margarine; milk (cow's milk) (all varieties) added to drinks; coffee (all varieties); milk (cow's milk) (all varieties) added to food; sugar (all varieties)

3.3.5 Associations of dietary patterns with BMI and presence of fatigue before joining SW (T0)

A dietary pattern score was created for each individual for each dietary pattern by using component scores. These scores were divided into quintiles to understand the relationship between these dietary patterns and presence of fatigue. Also, differences in the mean age and BMI for the lowest (quintile 1) and the highest (quintile 5) quintiles for the five dietary patterns at T0 were investigated by applying independent-samples t test or chi-square test.

The results indicated that BSW, participants in the highest quintile for the "savoury staples", "healthy snacks" and "milk and coffee" dietary patterns were significantly older than those in quintile one. In terms of BMI, participants in the highest quintile for the "CHO and high fat" and "meat and eggs" dietary patterns had significantly higher BMI than those in quintile one. Regarding fatigue, participants in the highest quintile for the "CHO and high fat" were more likely to have fatigue than those in quintile one ($p \leq 0.001$). There were no

significant differences in percentage of fatigue between quintiles one and five for the “savoury staples”, “healthy snacks”, “meat and eggs” and “milk and coffee” dietary patterns (table 3.6).

Table 3.6 Characteristics of participants with dietary pattern scores in the lowest (quintile 1) and in the highest (quintile 5) quintiles for the five dietary patterns at T0

	DP1: CHO and high fat		**DP2: Savoury staples**	
	Quintile 1	Quintile 5	Quintile 1	Quintile 5
Age (years)	84 | 37.73 (±8.9)	83 | 36.76 (±8.1)	**85 | 35.15 (±8.7) ***	**78 | 38.37 (±7.6) ***
BMI (kg/m^2)	**84 | 32.62 (±6.0) +**	**83 | 38.29 (±7.3) +**	86 | 35.87 (±7.3)	79 | 33.95 (±7.3)
Presence of fatigue (GFI≥28) n (%)	**41 (46.6) +**	**69 (77.5) +**	61 (69.3)	56 (62.9)

	DP3: Healthy snacks		**DP4: Meat and eggs**		**DP5: Milk and coffee**	
	Quintile 1	Quintile 5	Quintile 1	Quintile 5	Quintile 1	Quintile 5
Age (years)	**82 | 35.44 (±8.5) +**	**81 | 39.68 (±7.1) +**	**80 | 38.96 (±8.0) ***	**87 | 35.20 (±9.1) ***	**82 | 33.16 (±8.5) +**	**83 | 38.64 (±8.0) +**
BMI (kg/m^2)	84 | 36.08 (±8.1)	79 | 3 4.66 (±6.8)	**81 | 34.11 (±6.5) ***	**87 | 37.30 (±7.5) ***	83 | 36.89 (±8.2)	82 | 34.82 (±7.1)
Presence of fatigue (≥28) n (%)	54 (60.0)	56 (64.4)	55 (62.5)	55 (62.5)	56 (64.4)	59 (66.3)

* p≤0.05, + p≤0.001

Results expressed as number | mean (SD)

A multinomial logistic regression was used to analyse predictors for presence of fatigue. The reference category was "no fatigue" (GFI<28); "presence of fatigue" (GFI≥28) was compared to this reference group. The main interest of current analysis was focusing on the association of age and BMI with fatigue while controlling for dietary patterns. The results suggested that age and BMI had no significant effect on presence of fatigue at T0. The probability of fatigue was significantly increased by the "CHO and high fat" dietary pattern (OR=1.6, p=0.001). Other dietary patterns were not significant predictors of fatigue at T0 (table 3.7).

Table 3.7 Predictors of presence of fatigue (GFI≥28) at T0

Variables	**OR (95% CI)**	**SE**	**p value**
Age	1.01 (.98/1.04)	.01	0.62
BMI	1.02 (.98/1.05)	.02	0.21
Dietary Patterns	**OR (95% CI)**	**SE**	**p value**
CHO and high fat	1.59 (1.22/2.07)	.14	0.001
Savoury staples	.94 (.76/1.17)	.11	0.584
Healthy snacks	1.04 (.83/1.30)	.11	0.743
Meat and eggs	1.10 (.88/1.34)	.11	0.407
Milk and coffee	1.10 (.88/1.37)	.11	0.406

OR = Odds Ratio. SE = Standard Error. 95% CI = Confidence Interval.

3.3.5.1 Determination of dietary patterns from the FeFFQ for T1

The 35 most frequently consumed food groupings were identified for T1 (table 3.8) and subjected to PCA using SPSS. Same steps as determination of dietary

patterns at T0 were repeated for T1. Prior to performing PCA, the suitability of data for factor analysis was assessed and verified by the KMO value (0.730) and Barlett's Test p value (0.001).

PCA revealed that the presence of fourteen components with eigenvalue exceeding 1 (see appendix S). An inspection of the scree-plot revealed a clear break after the first component (see figure 3.3). However, parallel analysis showed eight components with eigenvalues exceeding the corresponding criterion values for a randomly generated data matrix of the same size (35 variables x 543 cases) (see appendix T). Because the results revealed from the PCA and parallel analysis regarding the optimal number of factors were inconsistent, the Component Matrix table was also interpreted (see appendix U). The table showed that majority of the items loaded quite strongly (above .4) on the first five components and it was decided to retain five components for further investigation.

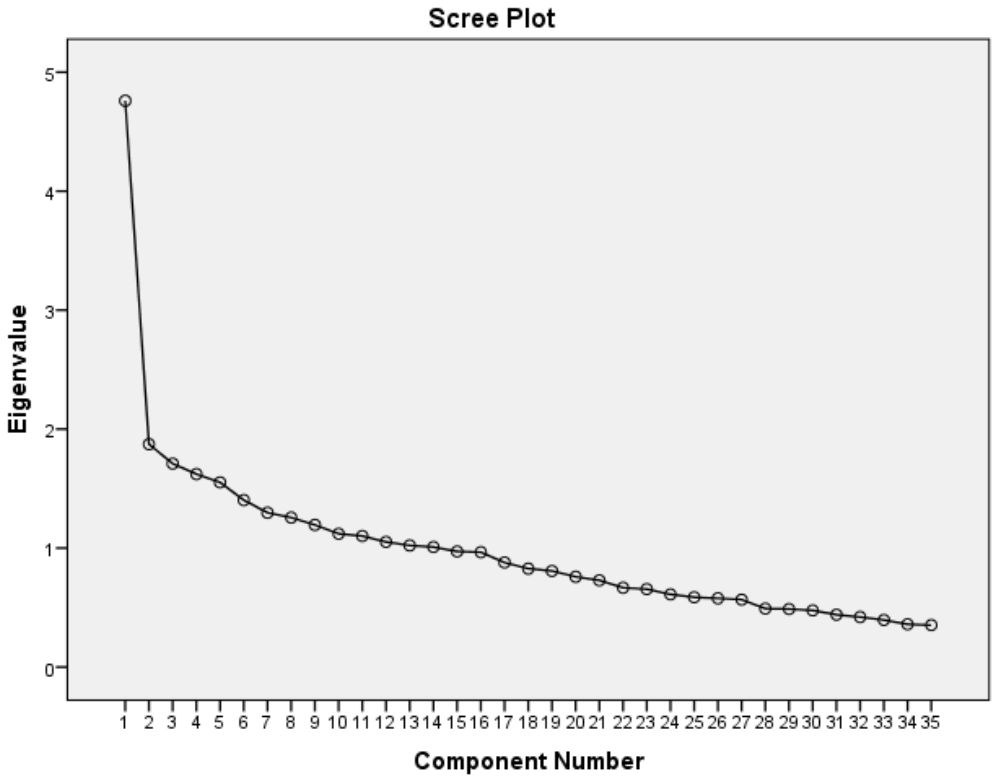
Table 3.8 The 35 most frequently consumed food groupings at T1

	Food groupings	Mean*	Med	SD	N	C
1	Milk (cow's milk) (all varieties) added to drinks	12.08	7	10.86	507	↓
2	Coffee (all varieties)	8.38	5	9.77	514	↑
3	Cordials (including syrups, powders)	6.53	2.5	9.01	509	↑
4	Apples	6.12	5	6.40	517	↑
5	Yoghurt	6.06	5	5.66	512	↑
6	Tomatoes (all varieties)	5.86	5	5.17	512	↑
7	Berries (e.g. strawberries, blackberries, cherries, blueberries, boysenberries, loganberries)	5.70	5	5.66	512	↑
8	Onions (all varieties), leeks, celery	5.25	5	4.07	516	↑
9	Citrus fruits	4.73	2.5	5.55	518	↑
10	Lettuce	4.63	5	4.21	515	↑
11	Bananas, green bananas	4.32	2.5	4.45	519	↑
12	Eggs	4.13	2.5	3.46	515	↑
13	Herbal tea, fruit tea	4.00	0.5	7.46	514	↑
14	Peppers (all varieties)	3.68	2.5	3.20	516	↑
15	Potato (eg. boiled, mashed, baked, roasted, fried, chips)	3.65	2.5	2.52	518	↓
16	Black tea	3.59	0	7.64	517	↓
17	Carrots	3.50	2.5	3.07	512	↑
18	Chicken, turkey or duck	3.40	2.5	2.74	500	↑
19	Mushrooms	3.19	2.5	3.03	514	↑
20	Courgette, zucchini, cucumber, gerkins or marrow (all varieties)	3.16	2.5	3.38	517	↑
21	Grapes	2.94	2.5	3.49	514	↑
22	Ham, bacon	2.67	2.5	2.88	505	↑
23	Broccoli (all varieties)	2.47	2.5	2.03	517	↑
24	Wholegrain bread and rolls	2.44	2.5	2.43	517	↑
25	Beans in sauce	2.04	2.5	1.98	515	↑
26	Green beans, broad beans, runner beans, asparagus	2.02	1	2.45	513	↑
27	Milk (cow's milk) (all varieties) added to food	2.01	0.5	3.29	511	↓
28	Stone fruit	1.87	1	2.86	515	↑
29	Peas, green	1.87	1	1.87	513	↑
30	Beetroot, radishes (all varieties)	1.85	1	2.80	518	↑
31	Beef	1.79	1	2.48	504	↑
32	Pasta, noodles (white)	1.68	1	1.73	511	↑
33	Porridge, rolled oats, oat bran, oat meal	1.68	0.5	2.22	515	↑
34	Spinach, silver beet, swiss chard (all varieties)	1.62	0.5	2.73	512	↑
35	Pears	1.60	0.5	2.79	511	↑

*The average weekly consumption of each food groupings.

Med, median; **C**, Change from T0 to T1; ↑, increase and ↓, decrease

Figure 3.4 Output from scree test at T1



3.3.5.1.1 Labelling the dietary patterns

The five-component solution explained a total of 32.9% of the variance, with Component 1 contributing 13.6%, Component 2 contributing 5.4%, Component 3 contributing 4.9%, Component 4 contributing 4.6% and Component 5 contributing 4.4%. To aid in the interpretation of these five components, varimax rotation was performed and five dietary patterns were labelled as follows:

1. **Vegetables:** Onions (all varieties), leeks, celery; beetroot, radishes (all varieties); lettuce; green beans, broad beans, runner beans, asparagus; tomatoes (all varieties); carrots; peppers (all varieties); courgette, zucchini, cucumber, gherkins or marrow (all varieties); broccoli (all varieties); spinach, silver beet, swiss chard (all varieties); peas, green; mushrooms

2. **Fruits and yoghurt:** Tomatoes (all varieties); grapes; citrus fruits; strawberries, blackberries, cherries, blueberries, boysenberries, loganberries, cranberries, gooseberries, raspberries; bananas, green bananas; pears; apples; stone fruit; yoghurt

3. **Healthy CHO and eggs:** Beans in sauce; potato; eggs; pasta, noodles (white); peas, green; mushrooms

4. **Milk, tea and coffee:** Milk (cow's milk) (all varieties) added to drinks; black tea; coffee (all varieties); milk (cow's milk) (all varieties) added to food

5. **Meat:** Chicken, turkey or duck; beef; ham, bacon; herbal tea, fruit tea

3.3.6 Associations of dietary patterns with BMI and presence of fatigue at T1

Analysis of independent-samples t test or chi-square test revealed that participants in the highest quintile for the "vegetables" and "milk, tea and coffee" dietary patterns were significantly older than those in quintile one.

However, participants in the highest quintile for the “meat” dietary pattern were significantly younger than those in quintile one. In terms of BMI, although participants in the highest quintile for the “healthy CHO and eggs” dietary pattern had significantly higher BMI than those in quintile one, participants in the highest quintile for the “milk, tea and coffee” dietary pattern had significantly lower BMI than those in quintile one. Regarding fatigue, participants in the highest quintile for the “vegetables” were less likely to have fatigue than those in quintile one (table 3.9).

Table 3.9 Characteristics of participants with dietary pattern scores in the lowest (quintile 1) and in the highest (quintile 5) quintiles for the five dietary patterns at T1

	DP1: Vegetables		**DP2: Fruits and yoghurt**	
	Quintile 1	Quintile 5	Quintile 1	Quintile 5
Age (years)	**79 | 35.61 (±8.9) ***	**78 | 38.78 (±7.9) ***	80 | 35.29 (±8.6)	80 | 37.21 (±8.0)
BMI (kg/m^2)	81 | 34.56 (±7.4)	78 | 33.22 (±6.5)	80 | 34.85 (±7.0)	80 | 34.77 (±6.5)
GFI Presence of fatigue (≥28) n (%)	**36 (44.4) ***	**20 (25.0) ***	30 (36.6)	27 (33.3)

	DP3: Healthy CHO and eggs		**DP4: Milk, tea and coffee**		**DP5: Meat**	
	Quintile 1	Quintile 5	Quintile 1	Quintile 5	Quintile 1	Quintile 5
Age (years)	73 | 37.81 (±8.2)	79 | 35.75 (±8.6)	**78 | 32.77 (±8.3) +**	**76 | 39.96 (±7.2) +**	**81 | 38.32 (±8.0) ***	**79 | 35.32 (±8.7) ***
BMI (kg/m^2)	**75 | 32.29 (±6.6) ***	**78 | 35.05 (±7.9) ***	**77 | 36.73 (±7.4) +**	**76 | 32.55 (±6.2) +**	81 | 34.23 (±7.0)	78 | 34.91 (±7.0)
GFI Presence of fatigue (≥28) n (%)	21 (26.2)	21 (25.9)	27 (33.3)	31 (37.8)	24 (28.6)	24 (28.9)

* p≤0.05, + p≤0.001

Results expressed as number | mean (SD)

Analysis of multinomial logistic regression suggested that higher BMI (OR=1.05) increased the probability of fatigue at T1 (p=0.05) and the probability of fatigue was significantly decreased by the "vegetables" dietary pattern (OR=.719, p=0.01). Other dietary patterns were not significant predictors of fatigue at T1 (table 3.10).

Table 3.10 Predictors of fatigue (GFI≥28) at T1

Variable	OR (95% CI)	SE	p value
Age	.994 (.97/1.02)	.02	0.695
BMI	1.05 (1.01/1.08)	.02	0.005
Dietary Patterns			
Vegetables	.719 (.56/.93)	.13	0.013
Fruits and yoghurt	1.08 (.86/1.34)	.11	0.518
Healthy CHO and eggs	.857 (.67/1.09)	.12	0.209
Milk, tea and coffee	1.24 (.96/1.60)	.13	0.095
Meat	.922 (.74/1.16)	.12	0.479

3.3.7 Determination of the most frequently consumed food groupings at T2

Similar as T0 and T1, the 35 most frequently consumed food groupings were identified for T2 (table 3.11). However, PCA was not used to identify dietary patterns at T2. There are two reasons for this. First, the sample size (n=74) for T2 was very small for factor analysis (Tabachnick & Fidell (2013) suggested at least 300 cases for factor analysis and Nunnally (1978) recommended ten cases for each variable to be factor analysed). Secondly, KMO analysis showed that the data set was not suitable for factor analysis (KMO measure of sampling adequacy= 0.503).

Table 3.11 The 35 most frequently consumed food groupings at T2

	Food groupings	Mean*	Med	SD	N	C
1	Milk (cow's milk) (all varieties) added to drinks	15.97	17.5	10.68	74	↑
2	Coffee (all varieties)	12.48	17.5	10.51	74	↑
3	Tomatoes (all varieties)	5.85	5	5.05	74	↓
4	Yoghurt	5.84	5	5.24	74	↓
5	Black tea	5.83	0	9.75	74	↓
6	Cordials (including syrups, powders)	5.55	2.5	8.20	74	↓
7	Apples	5.51	2.5	6.23	74	↓
8	Berries (e.g. strawberries, blackberries, cherries, blueberries, boysenberries, loganberries)	5.01	2.5	5.35	74	↓
9	Onions (all varieties), leeks, celery	4.80	5	3.98	74	↓
10	Citrus fruits	4.41	2.5	5.20	74	↓
11	Mushrooms	4.03	3.75	3.25	74	↑
12	Bananas, green bananas	3.82	2.5	4.28	74	↓
13	Carrots	3.51	2.5	3.31	74	↑
14	Peppers	3.29	2.5	3.79	74	↓
15	Eggs	3.25	2.5	2.66	74	↓
16	Herbal tea, fruit tea	3.21	0.5	6.36	74	↓
17	Potato	3.13	2.5	1.97	74	↓
18	Courgette, zucchini, cucumber, gherkins or marrow	2.90	2.5	2.82	74	↓
19	Chicken, turkey or duck	2.81	2.5	1.71	74	↓
20	Stone fruit	2.75	2.5	3.72	74	↑
21	Ham, bacon	2.66	2.5	3.49	74	↓
22	Mushrooms	2.61	2.5	2.12	74	↓
23	Wholegrain bread and rolls	2.61	2.5	2.74	74	↑
24	Milk (cow's milk) (all varieties) added to food	2.49	1	2.76	74	↑
25	Butter or margarine	2.47	1	2.96	74	↑
26	Grapes	2.29	1	3.20	74	↓
27	Broccoli (all varieties)	2.07	2.5	1.65	74	↓
28	Cooking oil (all varieties)	2.05	1	2.37	74	↑
29	Peas, green	1.99	1	1.80	74	↑
30	Green beans, broad beans, runner beans, asparagus	1.73	1	1.72	74	↓
31	Pears	1.70	0.5	2.87	74	↑
32	Beetroot, radishes	1.61	0.5	2.08	74	↓
33	Pawpaw (papaya), other melons	1.61	0.5	3.63	74	↑
34	Hard cheese	1.53	1	1.72	74	↑
35	Beef	1.47	1	1.32	74	↓

*The average weekly consumption of each food groupings. **Med**, median; **C**, Change from T1 to T2; ↑, increase and ↓, decrease

3.3.3 Dietary habits

Data from the dietary habits questionnaire showed that the percentage of participants who reported always (4 to7 times/week) having breakfast increased significantly from T0 to T1 (50.8% to 83.6%, $p<0.001$) with a small but significant decrease between T1 and T2 (83.6% to 80.6%, $p=0.001$). Similar to the findings for breakfast, there was a significant increase in the percentages of participants who reported always having lunch between T0 and T1 (71.6% to 89.7%, $p<0.001$) with a small decrease between T1 and T2 (89.7% to 86.1%, $p>0.05$). In terms of eating or drinking after evening meal the percentages decreased significantly from T0 to T1 (60.9% to 30.1%, $p<0.001$) with a small increase between T1 and T2 (30.1% to 44.3%, $p>0.05$).. Similarly, the percentages of participants who reported always having breakfast decreased significantly from T0 to T1 (20.9% to 8.4%, $p<0.001$) with a slight increase from T1 to T2 (8.4% to 10.3, $p>0.05$) (table 3.12).

Whilst around two-thirds of the participants (69.4%) reported they usually ate bread or toast at breakfast at T0, this reduced to 24.5% at T1 ($p<0.001$) with a small but significant increase from T1 to T2 (27.6% to 35.5% [n=74], $p<0.001$). A further change at breakfast was in fruit consumption. Between T0 and T1, there was nearly a four-fold increase in fruit intake (16.8% to 65.2%, $p<0.001$) with a small but significant decrease between T1 and T2 (68.4% to 63.2% [n=74], $p<0.001$) (table 3.13).

Table 3.12 Frequency of eating or drinking at the following time periods

	Always (4-7 times/week)	Sometimes (2-3 times/ week)	Never (0-1 time/week)
Before breakfast (%)			
T0 (n=543)	17.6	12.3	70.1
T1 (n=543)	19.6	7.2	73.2
T2 (n=74)	20.0	6.7	73.3
Breakfast (%)			
T0 (n=543)	50.8	27.5	21.7
T1 (n=543)	83.6	12.5	4.0
T2 (n=74)	80.6	18.1	1.4
Between breakfast and lunch (%)			
T0 (n=543)	47.3	37.5	15.2
T1 (n=543)	36.6	39.8	23.6
T2 (n=74)	47.1	32.4	20.6
Lunch (%)			
T0 (n=543)	71.6	22.9	5.5
T1 (n=543)	89.7	8.0	2.3
T2 (n=74)	86.1	13.9	0.0
Between lunch and evening meal (%)			
T0 (n=543)	55.6	31.5	12.8
T1 (n=543)	36.0	44.8	19.2
T2 (n=74)	40.6	42.0	17.4
Evening meal (%)			
T0 (n=543)	85.6	11.8	2.6
T1 (n=543)	90.1	7.6	2.3
T2 (n=74)	94.4	5.6	0.0
After evening meal (%)			
T0 (n=543)	60.9	30.4	8.7
T1 (n=543)	30.1	43.8	26.1
T2 (n=74)	44.3	42.9	12.9
Other e.g. midnight snack (%)			
T0 (n=543)	20.9	22.5	56.6
T1 (n=543)	8.4	10.6	81.1
T2 (n=74)	10.3	13.8	75.9

Table 3.13 Types of food eating during breakfast at T0, T1 and T2

	T0 (%)		T1 (%)		T2 (%)	p value	
	n=543	n=74	n=543	n=74	n=74	T0-T1 (n=543)	T1-T2 (n=74)
Breakfast cereals	51.9	61.8	39.2	55.3	56.6	<0.001	<0.001
Porridge	32.8	44.7	39.2	39.5	42.1	0.007	<0.001
Bread or toast	69.4	76.3	24.5	27.6	35.5	<0.001	<0.001
Noodles or rice	4.1	3.9	3.9	2.6	1.3	1.000	1.000
Milk products	41.6	50.0	50.8	64.5	43.4	<0.001	<0.001
Fruit	16.8	21.1	65.2	68.4	63.2	<0.001	<0.001
Meats	33.9	35.5	33.0	31.6	28.9	0.734	<0.001
Baked beans or eggs	30.2	32.9	51.2	55.3	36.8	<0.001	<0.001
Vegetables	6.3	7.9	17.1	18.4	13.2	<0.001	0.125

When the participants were asked whether they usually had a drink up to 1 hour before, with, or up to 1 hour after their breakfast, over 80% of the respondents reported ‘yes’ (82.2%, 87.5% and 94.7% at T0, T1 and T2 respectively) (table 3.14). The majority of the respondents reported that they usually drank coffee and tea over the three time points (T0, T1 and T2). Consumption of fruits and vegetable juices reduced significantly between T0 and T1 (18.6% to 3.1%, $p<0.001$) with a small increase at T2 (5.3%). However, there was a nearly double increase in water intake between T0 and T1 (26.2% to 49.0%, $p<0.001$) (table 3.15).

Table 3.14 Having a drink up to 1 hour before, with, or up to 1 hour after breakfast at T0, T1 and T2

T0 (%)		T1 (%)		T2 (%)	p value	
n=543	n=74	n=543	n=74	n=74	T0-T1 (n=543)	T1-T2 (n=74)
82.2	96.1	87.5	90.8	94.7	0.005	<0.001

Table 3.15 Types of drink consumed with breakfast at T0, T1 and T2

	T0 (%)		T1 (%)		T2 (%)	p value	
	n=543	n=74	n=543	n=74	n=74	T0-T1 (n=543)	T1-T2 (n=74)
Fruit or vegetable juices	18.6	17.1	3.1	2.6	5.3	<0.001	0.500
Milk or milk-based drinks (all varieties)	7.6	3.9	2.4	1.3	1.3	<0.001	-
Soy-based drinks	1.5	1.3	1.7	1.3	2.6	1.000	0.675
Chocolate-based drinks	4.2	0.0	0.6	0.0	0.0	<0.001	-
Coffee (all varieties)	43.8	52.6	38.5	48.7	51.3	<0.001	<.001
Tea (all varieties)	47.7	53.9	47.5	52.6	50.0	1.000	<.001
Water (all varieties)	26.2	28.9	49.0	50.0	44.7	<0.001	<.001
Other	10.7	7.9	9.8	9.2	6.6	0.441	0.688

Bread intake at lunch-time also decreased between T0 and T1 (86.4% to 11.8%, p<0.001) and slightly increased from T1 to T2 (10.5% to 27.6% [n=74], p=0.07). Fruit and vegetable intake increased (17.5% to 64.6%, p<0.001 [n=543], [fruit intake]; 32.2% to 82.9%, p<0.001 [n=543], [vegatable intake] from T0 to T1 respectively) (table 3.16). At all time-points, approximately 90% of the participants reported that they usually have a drink up to 1 hour before, with, or up to 1 hour after their lunch. Changes in the types of drinks consumed with

lunch was also similar to breakfast. Whilst intake fruit and vegetable juice and chocolate-based drinks decreased, water intake increased between T0 and T1 (table 3.17).

Table 3.16 Types of food eaten during lunch at T0, T1 and T2

	T0 (%)		T1 (%)		T2 (%)	p value	
	n=543	n=74	n=543	n=74	n=74	T0-T1 (n=543)	T1-T2 (n=74)
Bread	86.4	86.8	11.8	10.5	27.6	<0.001	0.07
Starchy food	45.5	39.5	48.8	48.7	36.8	0.243	<0.001
Milk products	33.0	39.5	37.0	42.1	25.0	0.109	0.05
Fruit	17.5	21.1	64.6	69.7	46.1	<0.001	<0.001
Meats	52.1	59.2	69.8	76.3	69.7	<0.001	<0.001
Legumes, nuts, eggs	12.3	18.4	28.9	35.5	21.1	<0.001	0.508
Vegetables	32.2	34.2	82.9	86.8	75.0	<0.001	<0.001

Table 3.17 Types of drink consumed with lunch

	T0 (%)		T1 (%)		T2 (%)	p value	
	n=543	n=74	n=543	n=74	n=74	T0-T1 (n=543)	T1-T2 (n=74)
Fruit or vegetable juices	10.3	9.2	2.4	3.9	3.9	<0.001	-
Milk or milk-based drinks (all varieties)	3.9	2.6	1.1	1.3	0.0	<0.001	<0.001
Soy-based drinks	0.4	0.0	0.4	0.0	0.0	-	-
Chocolate-based drinks	3.7	0.0	0.6	0.0	0.0	<0.001	-
Coffee (all varieties)	30.4	30.3	22.1	25.0	32.9	<0.001	<0.001
Tea (all varieties)	36.6	47.4	33.0	38.2	35.5	0.007	<0.001
Water (all varieties)	47.7	47.4	71.8	73.7	73.7	<0.001	-
Other	30.0	26.3	23.4	26.3	14.5	<0.001	0.065

At the evening meal, bread consumption again decreased over the three time points (33.3% to 4.2%, p<0.001[from T0 to T1] and 5.3% to 3.9%, p>0.05 [from T1 to T2]). However, fruit and vegetable intake increased (13.1% to 51.0%, p<0.001 [fruit intake]; 71.6% to 89.0%, p<0.001 [vegetable intake] from T0 to T1 respectively) (table 3.18). Over 90% of the participants reported that they usually have a drink up to 1 hour before, with, or up to 1 hour after their evening meal. Similar to breakfast and lunch, there was increase in water intake and decrease in consumption of fruit or vegetable juices and chocolate-based drinks. Also, there was almost a three-fold decrease in alcohol consumption between T0 and T1 (23.9% to 7.2%, p<0.001), but it significantly increased between T0 and T1 (13.2% to 21.1 [n=74], p≤.05) (table 3.19).

Table 3.18 Types of food eaten during evening meal

	T0 (%)		**T1 (%)**		**T2 (%)**	**p value**	
	n=543	**n=74**	**n=543**	**n=74**	**n=74**	**T0-T1 (n=543)**	**T1-T2 (n=74)**
Bread	33.3	34.2	4.2	5.3	3.9	<0.001	0.500
Starchy food	90.6	93.4	74.8	80.3	81.6	<0.001	<0.001
Milk products	39.2	34.2	25.0	28.9	19.7	<0.001	0.052
Meats	84.5	88.2	86.0	90.8	89.5	0.403	0.007
Legumes, nuts, eggs	24.1	27.6	34.8	43.4	26.3	<0.001	<0.001
Vegetables	71.6	80.3	89.0	97.4	90.8	<0.001	<0.001
Fruit	13.1	13.2	51.0	60.5	27.6	<0.001	<0.001

Table 3.19 Types of drink consumed with evening meal at T0, T1 and T2

	T0 (%)		T1 (%)		T2 (%)	p value	
	n=543	n=74	n=543	n=74	n=74	T0-T1 (n=543)	T1-T2 (n=74)
Fruit or vegetable juices	9.9	9.2	3.1	5.3	3.9	<0.001	0.748
Milk or milk-based drinks (all varieties)	4.8	3.9	1.3	1.3	1.3	<0.001	-
Soy-based drinks	0.6	0.0	0.4	0.0	0.0	1.000	-
Chocolate-based drinks	4.1	2.6	0.6	0.0	0.0	<.001	-
Coffee (all varieties)	16.9	22.4	12.7	17.1	17.1	<.001	-
Tea (all varieties)	31.5	40.8	29.1	35.5	32.9	0.106	<0.001
Alcohol	23.9	31.6	7.2	13.2	21.1	<0.001	0.027
Water (all varieties)	49.5	48.7	70.3	68.4	63.2	<0.001	<0.001
Other	35.0	31.6	31.1	30.3	27.6	<0.001	<0.001

Investigating the changes in food intake between meals, the most significant decreases from T0 to T1 were in biscuits or cakes (79.4%, 4.4% [p<.001]), potato crisps (77.9%, 6.8% [p<0.001]), and chocolate and sweets (79.6%, 6.1% [p<0.001]). However, intake of all of these foods significantly increased from T1 to T2 (p<0.001). The most significant increases were in fruit (28.0%, 84.2% [p<0.001]) and vegetables (4.6%, 44.9% [p<0.001] between T0 and T1 respectively). However, consumption of both fruit and vegetable significantly decreased between T1 and T2 (p<0.001) (table 3.20).

Table 3.20 Types of foods and drinks between meals at T0, T1 and T2

	T0 (%)		T1 (%)		T2 (%)	p value	
	n=543	n=74	n=543	n=74	n=74	T0-T1 (n=543)	T1-T2 (n=74)
Bread based snacks	34.1	27.6	0.9	0.0	2.6	<0.001	<0.001
Breakfast cereal	13.1	13.2	2.8	1.3	0.0	<0.001	<0.001
Porridge	3.1	1.3	2.4	1.3	0.0	0.503	<0.001
Biscuits or cakes	79.4	85.5	4.4	5.3	27.6	<0.001	<0.001
Crackers	26.9	34.2	6.1	3.9	10.5	<0.001	<0.001
Milk products	29.3	28.9	36.3	48.7	34.2	<0.001	<0.001
Fruit	28.0	31.6	84.2	92.1	85.5	<0.001	<0.001
Potato crisps	77.9	80.3	6.8	6.6	25.0	<0.001	<0.001
Cereal bars, muesli bars	29.8	26.3	22.5	23.7	31.6	<0.001	<0.001
Chocolate or sweets	79.6	78.9	6.1	3.9	21.1	<0.001	<0.001
Vegetables	4.6	3.9	44.9	50.0	22.4	<0.001	<0.001
Fruit or vegetable juices	9.9	7.9	7.0	3.9	5.3	0.065	0.500
Milk or milk-based drinks (all varieties)	7.9	3.9	2.2	0.0	1.3	<0.001	<0.001
Soy-based drinks	1.8	1.3	1.5	1.3	1.3	0.754	-
Chocolate-based drinks	14.9	11.8	2.0	0.0	3.9	<0.001	<0.001
Coffee (all varieties)	46.2	56.6	37.9	50.0	52.6	<0.001	<0.001
Tea (all varieties)	51.4	64.5	48.4	61.8	56.6	0.077	<0.001
Water (all varieties)	47.5	53.9	70.9	77.6	65.8	<0.001	<0.001
Other	21.9	22.4	19.2	23.7	23.7	0.054	-

When the participants were asked “During the entire week how many times did/do you eat out?”, the majority reported “once” and the percentage of those who selected this answer increased significantly from T0 to T1(30.0% to 44.9%, p<0.001) (table 3.21).

Table 3.21 Number of times eaten out during the entire week at T0, T1 and T2

	T0 (%)		T1 (%)		T2 (%)	p value	
	n=543	n=74	n=543	n=74	n=74	T0-T1 (n=543)	T1-T2 (n=74)
Never	14.4	23.7	35.4	42.1	27.6	<0.001	<0.001
Once	30.0	26.3	44.9	42.1	46.1	<0.001	<0.001
Twice	19.2	21.1	9.0	9.2	14.5	<0.001	0.050
Three times	19.9	14.5	3.3	3.9	6.6	<0.001	0.007
4 to 5 times	11.0	9.2	1.5	0.0	1.3	<0.001	<0.001
6 to 7 times	2.9	5.3	0.2	1.3	0.0	<0.001	<0.001
8 to 9 times	0.9	0.0	0.0	0.0	1.3	0.062	<0.001
10 to 11 times	0.0	0.0	0.0	0.0	0.0	-	-
12 to 13 times	0.0	0.0	0.0	0.0	0.0	-	-
14 plus times	0.4	0.0	0.0	0.0	0.0	0.500	-

Almost two-thirds of the participants (69.4%, 64.3% and 64.5%) at each time points indicated that when they were choosing foods and drinks, they did not know, or they did not consider whether foods have iron added to them. However, the percentages of participants who reported that they choose the product with added iron whenever they could, increased significantly from T0 to T1 (4.6% to 7.6%, p=0.002) (table 3.22).

Table 3.22 Frequency of choosing the product with added iron instead of the product without added iron

	T0 (%)		T1 (%)		T2 (%)	p value	
	n=543	n=74	n=543	n=74	n=74	T0-T1 (n=543)	T1-T2 (n=74)
Whenever I can	4.6	2.6	7.6	9.2	10.5	0.002	0.572
Usually	2.4	3.9	3.1	5.3	1.3	0.388	0.132
Sometimes	6.8	11.8	8.5	14.5	10.5	0.164	0.122
Never	15.5	14.5	11.4	7.9	9.2	<0.001	0.748
I don't know or I don't consider whether foods have iron added to them	69.4	67.1	64.3	63.2	64.5	<0.001	1.000

The participants reported that mean number of fruits and vegetables they ate per day increased significantly both during the week (fruits: 1.8 (±1.7) vs. 4.5 (±2.9), p<0.001; veg: 2.5 (±1.7), 4.8 (±2.5), p<0.001) and at the weekend (fruits: 1.4 (±1.5) vs. 4.0 (±2.0), p<0.001; veg: 2.2 (±1.8) vs. 4.3 (±1.9), p<0.001) from T0 to T1. Mean number of servings of meat, fish, chicken or seafood they consumed per day slightly increased between T0 and T1 both during the week (2.1 (±1.9) vs. 2.7 (±2.1), p<0.001) and at the weekend (2.00 (±1.3) vs. 2.28 (±1.4), p<0.001) (table 3.23).

Table 3.23 Mean number of servings of fruit, vegetables, and meat, fish, chicken or seafood per day during the week and at the weekend at T0, T1 and T2

	T0 (%)		T1 (%)		T2 (%)	p value
	n=543	n=74	n=543	n=74	n=74	T0-T1 (n=543)
Mean number of servings of **fruit** per day **during the week**	1.8 (±1.7) [0-12]	1.8 (±1.8) [0-11]	4.5 (±2.9) [0-15]	4.4 (±2.2) [0-10]	3.6 (±2.6) [0-15]	<0.001
Mean number of servings of **fruit** per day **at the weekend**	1.4 (±1.5) [0-12]	1.0 (±1.1) [0-5]	4.0 (±2.0) [0-14]	4.1 (±2.5) [0-14]	2.8 (±1.9) [0-10]	<0.001
Mean number of servings of **vegetables** per day **during the week**	2.5 (±1.7) [0-10]	2.6 (±1.6) [0-8]	4.8 (±2.5) [0-20]	4.7 (±2.3) [1-14]	3.6 (±2.2) [0-12]	<0.001
Mean number of servings of **vegetables** per day **at the weekend**	2.2 (±1.8) [0-20]	2.1 (±1.5) [0-6]	4.3 (±1.9) [0-12]	4.3 (±1.6) [1-10]	3.6 (±2.9) [0-22]	<0.001
Mean number of servings of **meat, fish, chicken or seafood per day during the week**	2.1 (±1.9) [0-10]	2.0 (±1.6) [0-10]	2.7 (±2.1) [0-12]	2.3 (±1.8) [0-10]	2.3 (±2.2) [0-12]	<0.001
Mean number of servings of **meat, fish, chicken or seafood per day at the weekend**	2.00 (±1.3) [0-10]	1.9 (±1.2) [0-8]	2.28 (±1.4) [0-10]	2.0 (±0.9) [0-4]	2.01 (±1.0) [0-6]	<0.001

3.3 SUMMARY AND DISCUSSION OF KEY FINDINGS

This study showed that approximately 40% of women reported experiencing low iron stores, ID or IDA before joining SW. However, only 9.6% of those experienced low iron stores, ID or IDA in the three months after becoming a member of SW.

The mean GFI of women before joining SW was 29.2 and the mean BMI=35.0 kg/m^2. In a previous study (Anandacoomarasamy *et al*., 2009) using the same fatigue questionnaire, it was found that the mean GFI was 24.9 in an obese population with a mean BMI=41.8 kg/m^2. In another study (Impellizzer *et al.*, 2013), the prevalence of fatigue in people who are obese (n=220, mean BMI=44.4 kg/m^2) was examined using the Fatigue Severity Scale and the prevalence was reported as 59%. Although a high level of fatigue in women who are obese might be related with ID, it also might be associated with excessive adipose tissue. Mehta (2015) reported that adiposity accelerates the development of stress-related neuromuscular fatigue. In an experimental study by Maffiuletti *et al.* (2007), muscle strength and fatigue were compared between individuals with obesity (n=10, mean BMI=41 kg/m^2) and healthy weight individuals (n=10, mean BMI=23 kg/m^2). Quantification of voluntary fatigue and relative strength indicated that quadriceps muscle function impairments were observed in people who are obese. It was suggested that this can contribute to the reduced functional capacity of individuals who are obese during daily-life activities and higher physiological demanding activities such as stair climbing and uphill walking.

In terms of the impact of the weight management programme on BMI, fatigue and PA levels in the 74 women for whom data was available over the three time-

points, the results show a statistically significant decrease in BMI and fatigue level, and a statistically significant increase in PA levels over the three time points. Studies investigating weight loss interventions that promote healthy eating and increasing PA similarly reported an increased PA during the intervention period (Jeffery et al., 2003; Lemstra et al., 2015; Wang et al., 2012).

The findings from the hierarchical multiple regression suggested that for the three control measures (baseline BMI, level of PA and decrease in level of fatigue), the highest beta value representing the unique contribution of each variable was PA level at T2 (beta=.54, $p < .005$). After controlling the effect of PA at T0, T1 and T2, baseline BMI and decrease in fatigue level explained only 9.5% of the variance in weight loss. But there was a significant positive correlation between decrease in GFI and weight loss. The relationship between PA, weight loss and fatigue can be explained by the effects of PA on iron status and low-grade inflammation in women who are obese. According to the systematic style reviews in this thesis, exercise is effective to improve iron status and reduce inflammation in women who are obese. Whilst women who feel less fatigued are likely to become more physically active, women who are more physically active might feel less fatigued. Further research is required to better understand the fatigue experience of women who are obese and the complex interaction between PA, fatigue and weight loss.

Analysis of FFQ showed that “milk added to drinks” and “coffee” were the two most frequently consumed food groupings at all the time periods (T0, T1 and T2). One of the most prominent findings from the FFQ was that mean frequency of yoghurt consumption increased from 2.37 to 6.06 from T0 to T1. Yoghurt can

be a good source of probiotics, when these have been added and nutrient-dense food improved gut health (Hussein *et al*., 2014) and improved gut health is effective to reduce inflammation in people who are obese (Xiao *et al*., 2014).

Regarding associations of dietary patterns with BMI, there was a positive association of "CHO and high fat" and "meat and eggs" dietary patterns with a higher BMI at T0 and "healthy CHO and eggs" dietary pattern with a higher BMI at T1. However, "milk, tea and coffee" dietary pattern was related to lower BMI at T1.

In terms of the relationship between the identified dietary patterns and presence of fatigue (GFI≥28), participants who adhere "CHO and high fat" dietary pattern were more likely to have fatigue at T0 and those who followed a "vegetable" dietary pattern were less likely to have fatigue at T1. Whilst energy-dense foods in "CHO and high fat" dietary pattern may lead to increase inflammation due to oxidative stress (Lüscher, 2015), the high content of dietary fibre and anti-oxidant in foods in "vegetable" dietary pattern might be related to reduced inflammation (Jiao *et al*., 2015; Chaiyasit *et al.*, 2005). One of the most frequent nonspecific symptoms of inflammation is fatigue and it is promoted by the increased production of acute-phase proteins and cytokines such as CRP, IL-6 and TNF-α in metabolic disorders (Lasselin & Capuron 2014). The inverse relationship between "vegetable" dietary pattern and level of fatigue may partly be explained by high content of prebiotic fibre in vegetables. Because prebiotic intake increases bioavailability of haem iron (Weinborn *et al*., 2017), it may lead to a reduction in fatigue level in iron deficient women who are obese.

With regard to dietary habits, one of the most prominent findings was that the percentages of participants who reported they always had breakfast increased significantly from T0 to T1. Breakfast cereals that are commonly enriched with iron was the most consumed food group after fruit at breakfast at both T1 and T2. According to data from the National Diet and Nutrition Survey (NDNS) Rolling Programme (years 5–6), breakfast cereals provided an average of 40% of the iron in the UK diet in adults (aged 19-64) (Bates *et al*., 2016) and breakfast cereals have a positive impact on iron status in women who are obese and had impaired iron status (Rodríguez-Rodríguez *et al*., 2007). Although the FFQ validated for iron-related dietary patterns and dietary habits questionnaire was designed to obtain data regarding iron-related eating habits, this study is limited by the absence a measure of dietary iron intake of women since the FFQ was not validated for it.

In terms of types of food consumed during main meals (breakfast, lunch and dinner), reported consumption of bread reduced and fruits and vegetables increased from T0 to T1. Regarding types of drinks consumed with meals, reported consumption of fruit juice decreased and water intake increased. With respect to the types of food and drinks consumed between meals and after evening meal, consumption of junk food including biscuits or cakes, potato crisps, chocolate or sweets decreased, and fruit and vegetable intake increased from T0 to T1. Whilst junk foods have a low iron content (Shewfelt, 2017), fruits and vegetables are good source of ascorbic acid which is one of the significant enhancers of the bioavailability of iron (Singh & Prasad, 2018). In addition, two systematic style reviews (chapter 2) suggested that reducing energy-dense foods by consuming more fibre and antioxidant rich foods such as fruits and vegetables

might be effective to both improve iron status and reduce inflammation in people who are obese.

A number of limitations need to be noted regarding the present study. First, this study was unable to assess participants' both iron related parameters and serum hepcidin levels. Although participants reported their ID, IDA or low iron stores status, it is possible that these results were biased due to the self-reported nature of the survey. Secondly, this study was limited by the absence of large number of participants at follow-up (T2). One reason for loss to follow-up might be related with no longer attending the SW group. Another reason was the lack of a technique to match responders between survey 1 and 2. Loss to follow up was a source of weakness in this study which could have affected the exploration of changes over the three time periods. Also, lack of sample size at T2 precluded to identify iron related dietary patterns via PCA. In conclusion, this chapter successfully demonstrated the interrelationship between BMI, fatigue level, dietary patterns and dietary factors affecting the bioavailability of iron and PA levels. SW is an effective weight management programme in helping to achieve reductions in BMI, fatigue levels and consumption of energy-dense foods, and increasing PA levels and consumption of high-fibre/antioxidant foods in non-pregnant women with obesity of child-bearing age. However, more research should be undertaken to explore the potential association between iron status, BMI, fatigue and motivation/commitment to lifestyle changes.

CHAPTER 4: AN INVESTIGATION OF THE RELATIONSHIP BETWEEN IRON AND WEIGHT STATUS IN WOMEN OF CHILD BEARING AGE IN TURKEY

The prevalence of both obesity and iron deficiency anaemia are relatively high in Turkish women (see sections 1.1.5.2 and 1.1.6.2 in chapter 1). Although the 3rd chapter successfully demonstrated the interrelationship between BMI, fatigue level, dietary patterns and dietary factors affecting the bioavailability of iron and PA levels, there is a need for a study on this topic that includes iron-related parameters before the association between ID and obesity is more clearly understood. The aim of this chapter is to describe the methods and results of the second study that aimed to identify any associations between BMI, iron status, fatigue level, dietary factors affecting the bioavailability of iron and PA level in Turkish women of child-bearing age who are obese using a paper-based survey. This was modelled on study 1, but with cultural/practical adjustments on the survey and availability of iron-related blood test results. Whilst the same questionnaires used in both Study 1 and 2 allow us to make a clear comparison between the UK and Turkey, iron-related blood parameters in this study contribute to a deeper understanding of the relationship between iron status, inflammation an obesity.

The research questions previously listed in section 1.2.4.3 (chapter 1) were investigated with the following objectives:

I. To determine mean BMI in Turkish women who consulted a registered dietitian in Turkey starting a healthy eating plan (T0) and 12 weeks after starting the plan (T1) and identify BMI change over these time periods.

II. To determine fatigue levels in women at T0 and T1 and identify changes over these time periods.

III. To determine PA levels in women at T0 and T1 and identify changes over these time periods.

IV. To explore iron status of women at baseline.

V. To determine association of iron status with BMI and level of fatigue.

VI. To identify the most frequently consumed food groupings in women at T0 and T1.

VII. To determine dietary habits in women at T0 and T1.

IX. To compare all available findings from UK and Turkey.

4.1 METHOD

4.1.1 Ethical considerations

Ethical approval for this follow-on study was obtained from Faculty of Medicine and Health Sciences, University of Nottingham (Ethics Reference No: OVS18102016) on 07/11/2016 (see appendix V). Then, permission was obtained from the General Secretariat of the Public Hospitals Union in Giresun and in Samsun, Turkey, in order to carry out the study in three state hospitals (one in Samsun and two in Giresun), on 23/12/2016 (see appendix W). The

hospitals were selected because both cities were medium-sized and transportation between the hospitals was convenient.

The participant information sheet, given to eligible women consulting a registered dietitian in Turkey, included the aim of the study, benefits and risks of taking part, the kinds of questions to be asked and confirmation that anonymity would be maintained at all times. Written consent was obtained for each woman who agreed to participate.

4.1.2 Study design

The researchers intended the study design to be as identical to study 1 as possible to allow for comparison. Hence the same questionnaire from study 1, but in paper-based format was used. This questionnaire was developed using questions from MAF (Belza *et al*., 1995), iron FFQ (Beck *et al.,* 2013), dietary habits questionnaire (Beck *et al.,* 2013) and IPAQ. The Turkish version of MAF and IPAQ short form were used and iron FFQ and dietary habits questionnaire were translated into Turkish.

All questionnaires were kept in the original translation and the Turkish language was used in the documentation given to the participants. However, some changes were made to the iron FFQ and dietary habits questionnaire to reflect Turkish eating habits. Firstly, pork and pork products including ham, bacon and black pudding were removed from the FFQ because of their unavailability in Turkey due to religious reasons. Secondly, ayran (traditional yogurt drink) and lahmacun (a thin dough baked with a topping of meat, tomato, and herbs) were added to the FFQ. Thirdly, although avocadoes and olives were in the same food groupings in the original questionnaire, they were separated in the Turkish

questionnaire because olives are highly consumed at the traditional Turkish breakfast, whilst avocados are consumed less frequently. On the dietary habits questionnaire, “olives” were added and “noodles or rice” were removed at the section of types of food eaten during breakfast. Also, “tea (all varieties)” were separated as “black tea (all varieties)” and “herbal/fruit tea (all varieties)” for all questions that included tea on the dietary habits questionnaire.

In addition to the first study data collection, baseline blood test results including glucose, triglyceride, haemoglobin, haematocrit, iron, iron binding capacity, ferritin, fasting insulin, and haemoglobin A1C (HbA1C) were recorded from the hospital records. The most recent blood test records (≤15 days before the survey date) were used.

4.1.3 Sampling

The study population was Turkish women of child-bearing age (19-49 years), not pregnant, who consulted a registered dietitian, at one of the three hospital out-patient clinics, to lose weight or receive a dietetic treatment for their chronic disease (eg. Type 2 diabetes) with a starting BMI ≥25 kg/m^2 over four weeks.

4.1.4 Data collection

After obtaining ethical approval, four registered dietitians from three different hospitals (one in Samsun and two in Giresun) were engaged, informed about the study and trained. Women who were referred to the Department of Nutrition and Dietetics in the state hospitals were informed about the study by the registered dietitians. Women who wished to participate in the study was given the participant information sheet and consent form.

Baseline weight (in kilogram) and height (in meter) were measured by the dietitians as part of the routine dietetic consultation. The questionnaire was completed by the participants just after their first visit to the dietitian in the waiting area. All documents belonging to the study were stored securely in the outpatient clinics of the dietitians.

The baseline survey (T0) was conducted in December 2016- January 2017. In April-May 2017, the second survey (T1) was posted to participants who provided their postal addresses in the first survey and wished to complete the second survey. The follow-up participants were asked to hand completed surveys in to their dietitian within 15 days after receiving it.

Before posting the second surveys, first surveys that included postal addresses were coded and the same codes were used for the second surveys in order to data match T0 and T1 responses.

4.1.5 Data Analysis

All survey data and the clinical data were collated and uploaded on the IBM SPSS version 24.0 for Windows (IBM Corp., Armonk, NY).

BMI was calculated by using measured weight and height at baseline. However, at follow-up, self-reported weight was used to calculate BMI, since the second surveys were not completed in the dietetic clinic. In terms of calculation of GFI and PA levels, the same procedures as the first study were repeated for baseline and follow-up (see sections 3.1.6.1.2 and 3.1.6.1.6 in chapter 3).

Due to the small sample size, dietary patterns from the FeFFQ could not be revealed by using factor analysis. However, the 40 most frequently consumed

food groupings based on the mean number of weekly consumptions of each food grouping were listed.

For the dietary habits questionnaire, exact McNemar's test was conducted using Stata Statistical Software Release 16 (StataCorp LLC., College Station, TX) with statistical significance assumed at $p \leq 0.05$.

Pearson product-moment correlation coefficient was used to investigate the association of iron status (as measured by Hb, haematocrit, iron, TIBC and ferritin) with obesity (as measured by BMI) and level of fatigue (as measured by GFI) at baseline.

4.2 RESULTS

The aim was to recruit approximately 100 participants to complete the first survey within a four-week period. Eighty-one eligible participants completed the first survey and 33 of those provided their postal addresses and reported that they wished to complete the second survey with 21 of those actually completing and returning the second survey. All participants who consented to take part in the study were included in the analysis. The baseline characteristics of all participants, and the baseline and follow-up characteristics of the participants who completed the both questionnaires are presented in table 4.1, 4.2 and 4.3 with statistical significance assumed at $p \leq 0.05$.

4.2.1 Sample description

The age range was 19-49 years with a mean age of 34 (±4.9) at baseline. The majority of women were non-smokers and were eating a variety of all foods including animal products. The majority of women had experienced two full-

term pregnancies. The majority of women were pre-menopausal and were having moderate period losses with a period duration of approximately seven days (table 4.1). The minimum time interval between T0 and T1 was 12 weeks and the maximum was 20 weeks.

Table 4.1 Characteristics of the study population at baseline in Study 2

	Baseline (n=81)
Mean age	34.0 (±9.6) [19-49]
Smoking status	
Yes	15.2%
No	73.4%
An ex-smoker	11.4%
Eating pattern	
Eat a variety of all foods, including animal products	88.2%
Eat eggs, dairy, fish and chicken but avoid other meat	3.9%
Eat eggs but avoid dairy products, all meats and fish (ovo-vegetarian	0.0%
Eat dairy products but avoid eggs, all meats and fish (lacto-vegetarian	2.6%
Eat eggs and dairy but avoid all meats and fish (ovo-lacto vegetarian)	2.6%
Eat no animal products including eggs, dairy, and honey (vegan)	0.0%
Eat fish or other sea food but avoid all other meats (pescatarian)	0.0%
Other	2.6%
Number of full-term pregnancies	1.85 (±1.3)
Postmenopausal status	
Yes	11.3%
No	88.8%
Mean period duration (day)	7.10 (±5.2) [0-30]
Volume of period	
Light	12.5%
Moderate	62.5%
Heavy	17.2%
Very heavy	7.8%

Although the majority of women were healthy with no/little chronic disease, nearly half of those (44.4%) reported that they had low iron stores, ID or IDA at baseline, and 43.2% of whom indicated that they received a treatment for ID at baseline. The percentage of women who had low iron stores, ID or IDA (in the last three months) and received a treatment for IDA (in the last three months) was 19.0% and 25.0% at follow-up, respectively (table 4.2).

Table 4.2 Acute or chronic illness, ID, IDA status of the study population

	T0		T1
Number	81	21	21
Acute or chronic illness status (%)			
Yes	34.2	38.1	19.0
No	59.5	47.6	81.0
Low iron stores, ID or IDA status (%)			
Yes	44.4	42.9	19.0
No	47.5	52.4	71.4
Receiving a treatment for ID (%)			
Yes	43.2	57.9	25.0
No	43.2	42.1	75.0

4.2.2 Changes in body weight, fatigue and physical activity levels from baseline to follow-up

A paired sample t-test was conducted to evaluate the impact of the dietetic treatment on women's BMI, GFI and PA level. There was a statistically significant decrease in BMI from T0 (31.4 (±5.6) kg/m^2) to T1 (29.1 (±5.2)

kg/m^2), ($p<0.001$). In terms of fatigue level, mean GFI was 29.2 (±11.4) at T0 and 27.7 (±11.1) at T1 ($p=0.578$) (table 4.3).

Analysis of IPAQ indicated that level of vigorous intensity PA increased from T0 (7.6 (±34.9) MET-min/week) to T1 (163.8 (±380.7) MET-min/week) with a trend towards significance ($p=0.078$). Similarly, there was an increase in level of moderate intensity PA from T0 (18.1 (±55.1) MET-min/week) to T1 (231.8 (±557.7) MET-min/week) showing no statistical significance ($p=0.090$). Walking MET-min per week was 526.0 (±541.8) at T0 and 476.5 (±464.2) at T1 ($p=0.759$). Overall, participants' total PA level was 551.8 (±525.4) MET-min/week at T0 and 872.2 (±774.0) MET-min/week at T1 ($p=0.155$) (table 4.3).

Table 4.3 Descriptive statistics for BMI, GFI and PA for T0 and T1

	Baseline (T0)		Follow-up (T1)	p-value
Number	81	21*	21	21
Mean age	34.0 (±9.6) [19-49]	34.0 (±6.5)	34.8 (±6.1) [24-49]	0.067
Mean BMI	32.4 (±4.9) [25.5-49.2]	31.4 (±5.6) [22.8-48.4]	29.1 (±5.2) [22.7-44.3]	<0.001
Mean GFI	28.1 (±11.2) [1.0-46.6]	29.2 (±11.4) [2.0-46.2]	27.7 (±11.1) [10.3-45.2]	0.578
Vigorous Intensity PA (MET-min/week)	123.5 (±529.9) [0.0-3600.0]	7.6 (±34.9) [0.0-160.0]	163.8 (±380.7) [0.0-1440.0]	0.078
Moderate Intensity PA (MET-min/week)	83.2 (±241.0) [0.0-1200.0]	18.1 (±55.1) [0.0-240.0]	231.8 (±557.7) [0.0-2400.0]	0.090
Walking (MET-min/week)	469.1 (±541.5) [0.0-2772.0]	526.0 (±541.8) [0.0-2079.0]	476.5 (±464.2) [0.0-1485.0]	0.759
Total PA (MET-min/week)	675.8 (±792.4) [0.0-3798.0]	551.8 (±525.4) [0.0-2079.0]	872.2 (±774.0) [0.0-3144.0]	0.155

*Statistical significance was determined for between T0 and T1 (n=21)

Note: The minimum time interval between T0 and T1 was 12 weeks and the maximum was 20 weeks.

4.2.3 Blood test results

Descriptive analysis of blood test results revealed that mean levels of glucose, haemoglobin, haematocrit, iron, iron binding capacity, ferritin and fasting insulin were in the reference range at baseline. The prevalence of IDA (Hb<12.0 g/dl) was 23.7% and low iron stores (ferritin <14 ng/ml) was 28.9%.

Mean triglyceride level (150.5 (±99.6) mg/dl) at baseline was above the reference range which may be a risk factor for atherosclerosis and non-alcoholic fatty liver disease. Also, mean HbA1C level (6.0 (±1.3) %) at baseline was higher than the normal range which reflects poorer control of diabetes and an indication of pre-diabetes. The percentage of participants who had higher HbA1c level than the reference range (>5.7%) was 40.0% (table 4.4).

Table 4.4 Mean blood test results of the study population and percentage of participants above or below the reference range at baseline

	Unit (reference range)	Percentage of participants above (A) or below (B) the reference range
Mean glucose (FBS) (n=77)		
100.6 (±28.9) [16.9-217.0]	mg/dl (70-110)	**A:** 19.5 % (n=15)
5.6 (±1.6) [0.9-12.1]	mmol/l (4.0-5.4)	
Mean triglyceride (n=67)		
150.5 (±99.6) [24.0-572.0]	mg/dl (50-150)	**A:** 35.8% (n=24)
1.7 (±1.3) [0.3-6.5]	mmol/l (0.3-1.8)	
Mean haemoglobin (n=76)		
12.8 (±1.1) [10.1-15.2]	g/dl (12.00-16.00)	**B:** 23.7% (n=18)
128 (±11.0) [101-152]	g/L (120-160)	
Mean haematocrit (n=76)		
39.7 (±6.2) [32.5-86.3]	% (36-46)	**B:** 11.8% (n=9)
Mean iron (n=34)		
68.0 (±44.9) [17.8-270]	mcg/dl (40-170)	**B:** 26.5% (n=9)
12.2 (±8.0) [3.2-48.3]	mcmol/L (10-28)	
Mean total iron-binding capacity (n=32)		
332.1 (±96.0) [27.5-500.0]	mcg/dl (250-450)	**A:** 9.4% (n=3)
59.4 (±17.2) [4.9-89.5]	mcmol/L (45-72)	
Mean ferritin (n=45)		
29.6 (±20.1) [4.1-88.1]	ng/ml (14-150)	**B:** 28.9% (n=13)
Mean fasting insulin (n=18)		
21.7 (±7.6) [11.2-38.0]	mIU/L (5-25)	**A:** 33.3% (n=6)
Mean HbA1C (n=30)		
6.0 (±1.3) [4.7-8.8]	% (3.5-5.7)	**A:** 40.0% (n=12)

4.2.4 Association of iron status with obesity and fatigue level

Pearson product-moment correlation coefficient was used to investigate the association of iron status (as measured by Hb, haematocrit, iron, TIBC and ferritin) with obesity (as measured by BMI) and level of fatigue (as measured by GFI). Whilst there was a small negative correlation between TIBC and BMI (r=-.138, n=32, p=0.45), a small positive correlation was found between ferritin and BMI (r=.148, n=45, p=0.33). In terms of level of fatigue, a small negative correlation was found between Hb and GFI (r=-.140, n=75, p=0.23) and ferritin and GFI (r=-.178, n=44, p=0.25) (table 4.5). However, all these correlations were small and not statistically significant (p>0.05).

Table 4.5 Correlation of iron status with BMI and GFI

IRON STATUS	BMI			GFI		
	r	n	p	r	n	p
Haemoglobin	-.076	76	0.51	-.140	75	0.23
Haematocrit	-.037	76	0.75	-.023	75	0.85
Iron	-.081	34	0.65	.084	34	0.64
TIBC	-.138	32	0.45	-.038	32	0.84
Ferritin	.148	45	0.33	-.178	44	0.25

4.2.5 Changes in the most frequently consumed food groupings from baseline to follow-up

The iron FFQ considered food consumption for the previous situation before starting a healthy eating plan (T0) and 12 weeks after starting the plan (T1). The average weekly consumption of each food grouping was determined, and the 40 most frequently consumed food groupings (as number of times consumed per week) at T0 and T1 were identified.

The results from FFQ indicated that "black tea" which is a potent inhibitor of intestinal absorption of non-haem iron was the most frequently consumed food both at T0 and T1 with a significantly decreased mean frequency of consumption from 15.9 (±9.6) to 12.5 (±10.6) (p=0.002). Altghough the most frequntly consumed food groupings were very similar at T0 and T1, mean frequency of intake of some food groupings changed from T0 to T1.

The most remarkable decreases in mean frequency from T0 to T1 were in "white bread and rolls" (11.8 to 4.4), "citrus fruits" (6.4 to 3.5), hard cheese (6.1 to 4.6), "butter or margarine" (5.9 to 4.2), "sugar" (8.2 to 1.9), "cracker" (2.4 to 0.9), "white rice" (2.3 to 1.2), "pears" (2.2 to 0.5), and "biscuits, chocolate or chocolate covered" (2.0 to 0.5). Some noticeable increases in mean frequency from T0 to T1 were in "yoghurt" (7.4 to 8.8), "soup, vegetable based, homemade" (6.4 to 7.7), "tomatoes" (6.1 to 8.2), "herbal tea, fruit tea" (4.5 to 5.9), "chicken, turkey or duck" (2.2 to 3.1), "beef" (1.9 to 4.2) and "dried apricots, prunes, dates, mixed dried fruit" (1.4 to 2.7) (table 4.6). No statistics were carried out on the changes in mean frequency of intake of food groupings from T0 to T1.

Table 4.6 The 40 most frequently consumed food groupings at T0 and T1

Time Period →	BASELINE (T0)												FOLLOW-UP (T1)					
Number →	81						21						21					
FOOD OR DRINK	O	N	Mean	Med	SD	Min-Max	O	N	Mean	Med	SD	Min-Max	O	N	Mean	Med	SD	Min-Max
Black tea	1	78	15.9	17.5	9.6	0-28	1	20	16.3	17.5	10.3	0-28	1	21	12.5	7	10.6	0-28
White bread and rolls	2	78	11.8	7	10.9	0-28	3	20	8.9	7.0	9.0	0-28	16	21	4.4	0.5	7.6	0-28
Cooking oil (all varieties)	3	74	10.5	7	8.1	0-28	2	18	12.9	7.0	9.5	1.0-28	4	21	8.7	7	7.6	0-28
Olives	4	74	8.5	7	7.3	0-28	7	18	7.9	7.0	7.6	0-28	2	18	9.8	7	8.3	0-28
Yoghurt	5	77	7.4	7	6.7	0-28	4	20	8.6	7.0	6.3	0.5-17.5	3	20	8.8	7	7.2	0-28
Wholegrain bread and rolls	6	78	7.4	3.75	8.9	0-28	8	20	7.7	5.0	9.1	0-28	5	21	8.4	7	7.0	0-17.5
Apples	7	74	6.8	5	7.0	0-28	5	19	8.1	2.5	8.3	0.5-28	12	18	5.0	3.75	5.2	0-17.5
Soup, vegetable based, homemade	8	78	6.4	5	5.8	0-28	6	20	8.0	7.0	6.9	0-28	7	21	7.7	7	5.2	0.5-17.5
Citrus fruits	9	76	6.4	5	6.4	0-28	9	20	7.7	5.0	6.9	0.5-17.5	21	19	3.5	1	5.3	0-17.5
Hard cheese	10	77	6.1	7	4.2	0-17.5	12	20	6.5	7.0	5.4	0-17.5	15	21	4.6	2.5	5.2	0-17.5
Tomatoes	11	77	6.1	5	6.8	0-28	13	20	6.1	6.0	5.6	0-17.5	6	21	8.2	7	7.3	0.5-28
Onions (all varieties), leeks, celery	12	76	6.0	6	5.7	0-28	14	19	5.7	5.0	6.8	0-28	10	20	5.6	5	5.8	0-17.5
Ayran	13	76	5.9	5	6.3	0-28	11	19	6.8	5.0	6.2	0.5-17.5	8	21	6.3	5	6.9	0-28
Butter or margarine	14	77	5.9	5	6.6	0-28	10	20	7.2	5.0	8.8	0-28	17	20	4.2	1.75	5.3	0-17.5
Sugar (all varieties)	15	73	5.6	2.5	8.2	0-28	17	17	4.4	1.0	7.6	0-28	37	19	1.9	0	4.4	0-17.5

Time Period →	BASELINE (T0)												FOLLOW-UP (T1)					
Number →	81						21						21					
FOOD OR DRINK	O	N	Mean	Med	SD	Min-Max	O	N	Mean	Med	SD	Min-Max	O	N	Mean	Med	SD	Min-Max
Coffee (all varieties)	16	76	4.8	2.5	6.1	0-28	18	19	4.3	2.5	5.2	0-17.5	13	20	4.8	5	4.2	0-17.5
Herbal tea, fruit tea	17	76	4.5	2.5	5.6	0-28	16	19	5.2	7.0	4.2	0-17.5	9	20	5.9	6	5.8	0-17.5
Lettuce	18	77	4.3	2.5	4.5	0-28	19	20	4.3	2.5	4.0	0-17.5	14	20	4.7	3.75	6.0	0-28
Eggs	19	78	4.3	5	3.0	0-17.5	21	20	4.2	3.8	2.6	0-7.0	11	21	5.1	7	3.9	0-17.5
Cottage cheese, ricotta cheese	20	77	3.8	2.5	3.4	0-17.5	25	19	3.4	2.5	3.1	0-7.0	25	20	3.0	1.75	4.3	0-17.5
Carrots	21	76	3.8	2.5	4.3	0-28	18	19	4.3	2.5	5.2	0-17.5	36	21	2.0	1	1.7	0-5
Other green leafy vegetables	22	77	3.4	2.5	4.0	0-28	23	21	3.8	1.0	6.2	0-28	19	19	4.1	1	7.1	0-28
Soup, meat based, homemade	23	75	3.4	1	5.5	0-28	20	19	4.2	5.0	4.6	0-17.5	20	21	3.5	1	4.3	0-17.5
Potato	24	78	3.1	2.5	2.9	0-17.5	30	21	2.9	2.5	2.2	0.5-7.0	23	19	3.2	2.5	4.0	0-17.5
Melons	25	74	2.5	0.5	3.7	0-17.5	26	18	3.2	1.0	4.6	0-17.5	31	19	2.2	0.5	4.2	0-17.5
Peppers (all varieties)	26	76	2.5	1	3.9	0-28	52	19	1.5	1.0	1.6	0-7.0	29	20	2.3	1	4.0	0-17.5
Crackers*	27	76	2.4	1	4.4	0-28	34	19	2.5	1.0	4.0	0-17.5	55	20	0.9	0.5	1.0	0-2.5
Watermelon	28	74	2.4	0.5	3.6	0-28	27	18	3.2	0.8	4.6	0-17.5	27	19	2.5	0.5	4.2	0-17.5
White rice*	29	77	2.3	2.5	2.9	0-17.5	31	19	2.9	2.5	3.8	0-17.5	47	21	1.2	1	1.0	0-2.5
Bananas	30	75	2.3	1	2.8	0-17.5	29	20	3.0	2.5	4.0	0-17.5	22	18	3.2	1.75	4.2	0-17.5
Stone fruit*	31	76	2.2	0.5	5.3	0-17.5	38	19	2.2	0.5	6.3	0-28	48	18	1.1	0.5	1.7	0-7
Chicken, turkey or duck	32	76	2.2	1	3.2	0-28	32	19	2.8	1.0	4.2	0-17.5	24	20	3.1	1	4.2	0-17.5
Pears*	33	74	2.2	1	4.1	0-17.5	22	18	3.9	1.0	7.3	0-28	70	18	0.5	0.5	0.6	0-2.5

Time Period →	BASELINE (T0)												FOLLOW-UP (T1)					
Number →	81						21						21					
FOOD OR DRINK	O	N	Mean	Med	SD	Min-Max	O	N	Mean	Med	SD	Min-Max	O	N	Mean	Med	SD	Min-Max
Peanuts, mixed nuts, pecan etc	34	78	2.1	1	2.7	0-28	37	20	2.2	2.5	1.6	0-7.0	30	21	2.3	1	2.6	0-7
Milk (cow's milk) as a drink	35	77	2.1	1	2.5	0-17.5	44	20	1.8	1.0	2.3	0-7.0	33	21	2.2	0.5	4.0	0-17.5
Lentils	36	77	2.1	1	3.3	0-7	28	20	3.1	1.8	6.0	0.5-28	35	21	2.0	1	1.8	0.5-7
Green beans, broad beans, runner beans, asparagus	37	76	2.0	1	2.4	0-28	39	19	2.0	2.5	1.9	0-7.0	32	20	2.2	1.75	2.0	0-7
Biscuits, chocolate or chocolate covered*	38	73	2.0	1	3.8	0-17.5	40	19	2.0	1.0	2.3	0-7.0	73	20	0.5	0.5	0.6	0-2.5
Beef	39	78	1.9	1	3.8	0-28	24	21	3.6	1.0	6.7	0-28	18	21	4.2	2.5	5.0	0.5-17.5
Red cabbage*	40	73	1.9	1	2.3	0-28	60	18	1.2	0.8	1.7	0-7.0	66	18	0.8	0	1.0	0-2.5
Dried apricots, prunes, dates, mixed dried fruit+	55	75	1.4	0.5	2.6	0-17.5	58	19	1.3	0.5	1.6	0-5	26	19	2.7	0.5	4.4	0-17.5
Strawberries, cherries+	50	75	1.5	0.5	2.7	0-17.5	53	19	1.5	0.5	2.2	0-7	28	20	2.5	1	2.4	0.5-7
Sultanas, raisins, currants, figs+	56	76	1.4	0.5	2.5	0-17.5	64	20	1.0	0.5	1.7	0-7	34	19	2.2	0.5	4.4	0-17.5
Peas+	54	76	1.4	1	3.3	0-28	33	19	2.6	1.0	6.3	0-28	38	20	1.6	1	1.7	0-7
Beans (canned or dried) +	53	78	1.4	1	3.2	0-28	36	20	2.4	1.0	6.1	0-28	39	20	1.5	1	1.7	0-7

Time Period →	BASELINE (T0)												FOLLOW-UP (T1)					
Number →	81						21						21					
FOOD OR DRINK	O	N	Mean	Med	SD	Min-Max	O	N	Mean	Med	SD	Min-Max	O	N	Mean	Med	SD	Min-Max
Almonds+	75	72	0.9	0.5	1.5	0-7	68	17	0.9	0.5	1.0	0-2.5	40	19	1.5	0.5	2.5	0-7

* Not in the 40 most frequently consumed food groupings list for T1 but in the list for T0

+ Not in the 40 most frequently consumed food groupings list for T0 but in the list for T1

O, order; **N**, number; **med**, median; **min-max**, minimum-maximum

4.2.6 Dietary Habits

Data from the dietary habits questionnaire indicated that the percentage of participants who reported always (4 to7 times/week) having breakfast decreased from 91.7% to 80.0% from T0 to T1 and those who reported never (0-1 time/week) having breakfast increased from 1.4% to 5.0%. Similar to the findings for breakfast, there was an increase in the percentages of participants who reported never having lunch and evening meal increased from T0 to T1 (lunch: 7.1% to 15.0%, evening meal: 1.4% to 5.0%). However, the percentage of participants who reported always eating or drinking between breakfast and lunch, increased from T0 to T1 (17.9% to 35.0%) (table 4.7). No statistics were perfomed on the changes in the frequency of eating or drinking at the periods in table 4.7.

Table 4.7 Frequency of eating or drinking at the following time periods at T0 and T1

	Always (4-7 times/week)	Sometimes (2-3 times/ week)	Never (0-1 time/week)
Before breakfast (%)			
T0 (n=81)	13.4	13.4	73.1
T1 (n=21)	17.6	11.8	70.6
Breakfast (%)			
T0 (n=81)	91.7	6.9	1.4
T1 (n=21)	80.0	15.0	5.0
Between breakfast and lunch (%)			
T0 (n=81)	17.9	40.3	41.8
T1 (n=21)	35.0	45.0	20.0
Lunch (%)			
T0 (n=81)	72.9	20.0	7.1
T1 (n=21)	65.0	20.0	15.0
Between lunch and evening meal (%)			
T0 (n=81)	27.5	40.6	31.9
T1 (n=21)	31.6	52.6	15.8
Evening meal (%)			
T0 (n=81)	95.7	2.9	1.4
T1 (n=21)	90.0	5.0	5.0
After evening meal (%)			
T0 (n=81)	40.0	31.4	28.6
T1 (n=21)	36.8	42.1	21.1
Other eg. Midnight snack (%)			
T0 (n=81)	7.8	18.8	73.4
T1 (n=21)	5.9	23.5	70.6

At the breakfast, although there was a decrease in eating bread or toast (82.1% to 50.0%, p=0.002) and meats (21.8% to 5.0%, p=0.001), an increase found in consumption of fruits (4.8% to 15.0%, p>0.05) and vegetables (66.7% to 80.0, p<0.001) from T0 to T1 (table 4.8). Over two-thirds of the participants reported they usually had a drink with their breakfast at T0 and T1 (table 4.9) and black tea was the most consumed drink with breakfast both at T0and T1 (52.4% [n=21]). The percentage of participants who reported drinking usually milk or milk-based drinks at breakfast was 4.8% at T0 and 0.0% at T1 (p=0.968). Those who drank herbal/fruit tea remained the same between T0 and T1 (14.3% [n=21]) (table 4.10).

Table 4.8 Types of food eaten during breakfast at T0 and T1

	T0 (%)		T1 (%)	p value
	n=81	n=21	n=21	T0-T1 (n=21)
Breakfast cereals	1.3	0.0	0.0	-
Porridge	1.3	0.0	0.0	-
Bread or toast	82.1	85.7	50.0	0.002
Milk products	71.8	71.4	70.0	<0.001
Fruit	5.1	4.8	15.0	0.727
Meats	21.8	23.8	5.0	0.001
Baked beans or eggs	87.2	85.7	90.0	<0.001
Vegetables	60.3	66.7	80.0	<0.001
Olives	87.2	71.4	95.0	0.125

Table 4.9 Having a drink up to 1 hour before, with, or up to 1 hour after breakfast

T0		T1	p value
n=81	n=21	n=21	T0-T1 (n=21)
71.8	71.4	71.4	-

Table 4.10 Types of drink consumed with breakfast

	T0 (%)		T1 (%)	p value
	n=81	n=21	n=21	T0-T1 (n=21)
Fruit or vegetable juices	2.6	0.0	4.8	0.968
Milk or milk-based drinks (all varieties)	6.5	4.8	0.0	0.968
Soy-based drinks	0.0	0.0	0.0	-
Chocolate-based drinks	1.3	0.0	0.0	-
Coffee (all varieties)	20.8	19.0	19.0	-
Black tea (all varieties)	48.1	52.4	52.4	-
Herbal/fruit tea (all varieties)	7.8	14.3	14.3	-
Water (all varieties)	48.1	42.9	47.6	0.982
Other	2.6	4.8	9.5	0.972

At lunch-time, starchy food intake decreased between T0 and T1 (47.6% to 19.0%, p=0.031). There was a slight increase in milk products intake over the same period (47.6% to 52.4%) showing no statistical significance (p=0.968) (table 4.11). Approximately three-quarters of the women reported that they usually have a drink up to 1 hour before, with, or up to 1 hour after their lunch both at T0 and T1. Regarding the types of drinks consumed with lunch, whilst intake of fruit or vegetable juices decreased (9.5% to 0.0%, p=0.500),

herbal/fruit tea intake increased (9.5% to 30.0%, p=0.062) between T0 and T1. However, these changes were not statistically significant (table 4.12).

Table 4.11 Types of food eaten during lunch time

	T0 (%)		T1 (%)	p value
	n=81	n=21	n=21	T0-T1 (n=21)
Bread	75.3	66.7	38.1	0.070
Starchy food	45.5	47.6	19.0	0.031
Milk products	45.5	47.6	52.4	0.967
Fruit	13.4	9.5	14.3	0.962
Meats	44.2	52.4	42.9	0.727
Legumes, nuts, eggs	45.5	47.6	42.9	0.982
Vegetables	79.2	71.4	71.4	-

Table 4.12 Types of drink consumed with lunch

	T0 (%)		T1 (%)	p value
	n=81	n=21	n=21	T0-T1 (n=21)
Fruit or vegetable juices	9.1	9.5	0.0	0.500
Milk or milk-based drinks (all varieties)	10.4	0.0	10.0	0.500
Soy-based drinks	0.0	0.0	0.0	-
Chocolate-based drinks	1.3	4.8	0.0	0.968
Coffee (all varieties)	28.6	42.9	30.0	0.453
Black tea (all varieties)	46.8	57.1	45.0	0.375
Herbal/fruit tea (all varieties)	13.0	9.5	30.0	0.062
Water (all varieties)	58.2	57.2	65.0	0.688
Other	6.5	4.8	5.0	0.987

At the evening meal, consumption of bread, starchy food, meats, legumes, nuts and eggs decreased, and vegetables intake slightly increased from T0 to T1 (table 4.13). Over 70% of the participants reported that they usually have a drink up to 1 hour before, with, or up to 1 hour after their evening meal at T0 and T1. Similar to breakfast and lunch, black tea was the most consumed drink after water. There was a decrease in fruit or vegetable juices and increase in milk or milk-based drinks, herbal/fruit tea and water between T0 and T1 (table 4.14).

Table 4.13 Types of food eaten during evening meal

	T0 (%)		T1 (%)	p value
	n=543	n=74	(n=21)	T0-T1 (n=543)
Bread	74.0	66.7	61.9	0.976
Starchy food	59.7	66.7	38.1	0.070
Milk products	49.4	57.1	52.4	0.978
Fruit	15.6	4.8	14.3	0.500
Meats	77.9	76.2	57.1	0.289
Legumes, nuts, eggs	68.8	71.4	57.1	0.453
Vegetables	85.7	81.0	90.5	0.625

Investigating the changes in food and drink intake between meals and after evening meal, similar food/drink groups increased (vegetables, herbal/fruit tea, and water) or decreased (bread-based snacks, biscuits or cakes, crackers, potato crisps, chocolate or sweets, and chocolate-based drinks) between T0 and T1 (table 4.15 and 4.16). However, whilst there was an increase in milk products and fruit and vegetable juices intake between meals (table 4.15), consumption of these food/drink groups decreased after evening meal from T0 to T1 (table

4.16). Also, although milk or milk-based drinks intake decreased between meals (table 4.15), it increased after evening meal (table 4.16).

Table 4.14 Types of drink consumed with evening meal

	T0 (%)		T1 (%)	p value
	n=81	n=21	n=21	T0-T1 (n=21)
Fruit or vegetable juices	6.5	4.8	4.8	-
Milk or milk-based drinks (all varieties)	10.4	9.5	14.3	0.962
Soy-based drinks	0.0	0.0	0.0	-
Chocolate-based drinks	1.3	0.0	0.0	-
Coffee (all varieties)	20.8	14.3	23.8	0.625
Black tea (all varieties)	55.8	57.1	57.1	-
Herbal/fruit tea (all varieties)	6.5	0.0	19.0	0.125
Alcohol	0.0	0.0	0.0	-
Water (all varieties)	70.4	69.4	81.0	0.070
Other	7.8	9.5	4.8	0.972

Table 4.15 Types of foods and drinks between meals

	T0 (%)		T1 (%)	p value
	n=81	n=21	n=21	T0-T1 (n=21)
Bread based snacks	15.4	9.5	4.8	0.972
Breakfast cereal	2.6	4.8	0.0	0.968
Porridge	1.3	0.0	4.8	0.968
Biscuits or cakes	47.4	47.6	4.8	0.004
Crackers	34.6	38.1	28.6	0.754
Milk products	25.6	19.0	33.3	0.508
Fruit	60.3	52.4	61.9	0.727
Potato crisps	16.7	4.8	4.8	-
Cereal bars, muesli bars	3.8	0.0	4.8	0.968
Chocolate or sweets	29.5	33.3	14.3	0.031
Vegetables	3.8	4.8	9.5	0.625
Fruit or vegetable juices	14.1	0.0	23.8	0.500
Milk or milk-based drinks (all varieties)	11.5	0.0	0.0	-
Soy-based drinks	1.3	0.0	0.0	-
Chocolate-based drinks	7.7	4.8	0.0	0.968
Coffee (all varieties)	37.2	33.3	33.3	-
Black tea (all varieties)	60.3	71.4	76.2	0.975
Herbal/fruit tea (all varieties)	26.9	23.8	33.3	0.688
Water (all varieties)	70.5	80.9	85.7	0.125
Other	5.1	4.8	14.3	0.500

Table 4.16 Types of foods and drinks after evening meal

	T0 (%)		T1 (%)	p value
	n=81	n=21	n=21	T0-T1 (n=21)
Bread based snacks	14.1	9.5	0.0	0.500
Breakfast cereal	2.6	4.8	0.0	0.968
Porridge	2.6	0.0	0.0	-
Biscuits or cakes	39.7	42.9	4.8	0.008
Crackers	30.8	38.1	4.8	0.039
Milk products	21.8	14.3	14.3	-
Fruit	52.6	61.9	47.6	0.508
Potato crisps	16.7	19.0	0.0	0.125
Cereal bars, muesli bars	2.6	0.0	0.0	-
Chocolate or sweets	28.2	38.1	14.3	0.125
Vegetables	3.8	0.0	4.8	0.968
Fruit or vegetable juices	12.8	14.3	4.8	0.625
Milk or milk-based drinks (all varieties)	9.0	19.0	19.0	-
Soy-based drinks	0.0	0.0	0.0	-
Chocolate-based drinks	3.8	0.0	0.0	-
Coffee (all varieties)	29.5	33.3	23.8	0.727
Black tea (all varieties)	61.5	61.9	57.1	0.957
Herbal/fruit tea (all varieties)	14.1	4.8	19.0	0.250
Alcohol	0.0	0.0	0.0	-
Water (all varieties)	65.3	61.9	76.2	0.180
Other	2.6	0.0	9.5	0.500

When the participants were asked “During the entire week how many times did/do you eat out?”, the majority reported “never” and the percentage of those who selected this answer decreased between T0 and T1 (25.0% to 14.3%, $p>0.05$) (table 4.17).

Table 4.17 Number of times eaten out during the entire week

	T0 (%)		T1 (%)	p value
	n=81	n=21	n=21	T0-T1 (n=21)
Never	47.4	25.0	14.3	0.625
Once	26.3	50.0	38.1	0.727
Twice	10.5	15.0	19.0	0.982
Three times	3.9	5.0	23.8	0.375
4 to 5 times	9.2	5.0	4.8	0.990
6 to 7 times	1.3	0.0	0.0	-
8 to 9 times	0.0	0.0	0.0	-
010 to 11 times	0.0	0.0	0.0	-
12 to 13 times	0.0	0.0	0.0	-
14 plus times	1.3	0.0	0.0	-

Almost half of the participants at T0 and T1 indicated that when they were choosing foods and drinks, they never choose the product with added iron (table 4.18).

Table 4.18 Frequency of choosing the product with added iron instead of a product without added iron

	T0 (%)		T1 (%)	p value
	n=81	n=21	n=21	T0-T1 (n=21)
Whenever I can	5.3	9.5	15.0	0.970
Usually	6.6	9.5	0.0	0.500
Sometimes	7.9	14.3	10.0	0.961
Never	44.7	33.3	50.0	0.508
I don't know or I don't consider whether foods have iron added to them	35.5	33.3	25.0	0.969

The participants reported that mean servings of fruits, vegetables, meat, fish, and chicken or seafood they ate per day increased between T0 and T1 both during the week and at the weekend (table 4.19).

Table 4.19 Mean number of servings of fruit, vegetables, and meat, fish, chicken or seafood per day during the week and at the weekend

	T0		T1	p value
	n=81	**n=21**	**n=21**	**T0-T1 (n=21)**
Mean number of servings of **fruit** per day **during the week**	1.97 (±1.2) [0-6]	1.55 (±1.2) [0-5]	2.37 (±1.5) [0.5-5.0]	0.034
Mean number of servings of **fruit** per day **at the weekend**	1.66 (±1.2) [0-5]	1.50 (±1.2) [0-5]	1.75 (±1.1) [0-4]	0.331
Mean number of servings of **vegetables** per day **during the week**	1.79 (±1.2) [0-5]	1.71 (±1.3) [0-5]	2.79 (±2.0) [0-8]	0.063
Mean number of servings of **vegetables** per day **at the weekend**	1.49 (±1.1) [0-5]	1.43 (±1.1) [0-4]	1.50 (±1.9) [0-12]	0.985
Mean number of servings of **meat, fish, chicken or seafood** per day **during the week**	1.14 (±1.0) [0-5]	1.20 (±0.8) [0-3]	1.55 (±1.4) [0-4]	0.002
Mean number of servings of **meat, fish, chicken or seafood** per day **at the weekend**	1.00 (±1.0) [0-4]	1.26 (±0.7) [0-3]	1.29 (±1.0) [0-3]	0.298

4.3 SUMMARY AND DISCUSSION OF KEY FINDINGS

Approximately half of the women reported that they, in the past, had had low iron stores, ID or IDA prior to consulting a dietitian (at baseline). Nearly one fifth of women reported that they had low iron stores, ID or IDA in the three months after consulting a dietitian. These results were consistent with the blood test results, which indicated that IDA was prevalent in 23.7% of the women and 28.9% of those had low iron stores. In this study, mean Hb concentration (g/l) was 128 (±11.0). These results reflect those of WHO (2015) which reported the prevalence of IDA in non-pregnant women in Turkey as 29.0% and mean Hb concentration in the same group as 127 g/l.. Because the report (WHO 2015) represents the overall population of non-pregnant women, it is encouraging to compare our findings with the population of non-pregnant women who are overweight or obese. Data from the 1999 Mexican Nutrition Survey (Cepeda-Lopez *et al*., 2011) indicated that the prevalence of ID among non-pregnant women who are overweight (n=224, BMI 25-30 kg/m^2) was 52.7% and 61.2% among non-pregnant women who are obese (n=157, BMI ≥30 kg/m^2). These rather contradictory results may be explained by the fact that whilst in our study the prevalence of IDA (Hb<12.0 g/dl) was investigated, (Cepeda-Lopez *et al*., 2011) investigated the prevalence of ID that was defined as either low serum iron (<60 µg/dl) or elevated TIBC (>360 µg/dl) and low transferrin saturation (<20%) values. Mean Hb concentrations (g/l) in women who are overweight and obese were 135 (±16.0) and 137 (±16.0), respectively and both values higher than those we observed (128 (±11.0)).

In terms of changes in BMI, fatigue and PA level, there was a statistically significant decrease in BMI, a decrease in GFI, and an increase in PA level from baseline to follow-up.

With regards to the relationship between iron status and fatigue level and BMI, GFI was negatively correlated with both Hb and ferritin. There was an inverse correlation between TIBC and BMI, and a positive correlation between ferritin and BMI. These results accord with our earlier observations, which showed a negative correlation between TIBC and BMI (Cepeda-Lopez *et al*., 2015) and a positive correlation between ferritin and BMI (Khan *et al*., 2016). These relationships may partly be explained by the fact that elevated serum ferritin and reduced TIBC levels in response to anaemia of chronic disease/inflammatory response although reduced serum ferritin and elevated TIBC is expected for ID (Kelly *et al*. 2017). These results confirm the association between obesity and ID dependent of low-grade inflammation.

Since this study included women consulting a dietitian for not only weight management but also dietetic treatment for obesity related chronic diseases such as type 2 diabetes, 40% of women had an elevated HbA1C level and one-third of women had elevated fasting insulin levels. One characteristic of type 2 diabetes is chronic inflammation and it contributes to fatigue symptoms in patients with type 2 diabetes (Lasselin *et al*., 2012). Fatigue level could be decreased by increased PA level. The systematic style reviews in this thesis suggested that PA might improve iron status and inflammation in women who are obese. A broadly similar point was also made by El-Kader *et al*. (2015), who suggested that treadmill walking exercise training is effective in reducing

inflammation-related fatigue symptoms in people with type 2 diabetes and obesity. Another way of reducing inflammation might be increasing prebiotics and/or probiotics content of the diet. Cani *et al.* (2012) demonstrated that gut microbiota influences energy balance, glucose metabolism and low-grade inflammation associated with obesity and related metabolic disorders.

The analysis of the FFQ showed that "black tea" was the most consumed food grouping both at T0 and T1. Whilst the mean frequency of consumption of "white bread and rolls", "butter and margarine", "sugar" and "biscuits, chocolate or chocolate covered" decreased, consumption of "yoghurt", "herbal tea, fruit tea", "chicken, turkey or duck" and "beef" increased. A decrease in the consumption of "citrus fruits" and "pears" from baseline to follow up might be related with their seasonal availability. The first survey was conducted between December and January which is the season of "citrus fruits" and "pears" are highly available in Turkey. However, in April (month of the second survey), their availability reduces. Since the study was limited to small sample size, it was not possible to identify the iron-related dietary patterns of participants. However, a decrease in the consumption of energy-dense foods is very clear.

The dietary habits questionnaire revealed that the percentage of participants who reported always eating or drinking between breakfast and lunch, increased from T0 to T1. This finding supports the advice commonly given by Turkish dietitians; "eat little and often". A possible explanation for the achieved weight loss might be restricting intake at main meals and having smaller snacks instead with a decrease in total energy and increase in fruits and vegetables intake. Consumption of bread, toast or starchy food decreased at breakfast, lunch and

evening meal from baseline to follow-up and, and vegetable increased from baseline to follow-up.

The majority of the participants reported that they usually have a drink up to 1 hour before, with, or up to 1 hour after their breakfast, lunch and evening meal. Black tea was the most consumed drink after water both T0 and T1.

The most prominent finding from both FFQ and dietary habits questionnaire was that women consumed black tea and usually with meals. Although iron status in Western populations in which most people have adequate iron stores (serum ferritin) was not affected by tea consumption (Temme & Van Hoydonck, 2002), high consumption of black tea with a meal might be one of the most important risk factor for ID in women in Turkey where the prevalence of both ID and obesity is high. Another finding from both FFQ and dietary habits questionnaire was that women increased consumption of herbal/fruit tea from baseline to follow-up. This seems to be a useful strategy to reduce the risk of ID. However, more recently, a cross-sectional study (Mennen *et al*., 2007) found that serum ferritin concentration was not related to type of tea (black, green, or herbal), strength of tea, time of tea drinking and iron status of individual in a French population (n=2593). Since studies undertaken so far provide conflicting evidence, more studies will be needed to develop a full picture of the impact of tea consumption on iron absorption and status.

The generalisability of these results is subject to certain limitations. Firstly, the study did not assess participants' serum hepcidin level, which is influenced by low-grade inflammation due to excess adiposity and reduces iron absorption. Secondly, in spite of the availability of iron related parameters at baseline in this

study, it was unfortunate that the study did not include the blood test results at follow-up. Thirdly, interpretation of weight loss between baseline and follow-up were potentially biased because of the ranging time interval from 12 to 20 weeks. Also, the small sample size limits the generalisability of its findings. In conclusion, this chapter further supported the interrelationship between BMI, fatigue level, dietary patterns and dietary factors affecting the bioavailability of iron and PA levels. Also, in addition to the Study 1, the relationship between iron status and low-grade inflammation in women who are obese was revealed. Although BMI and fatigue level of women in this study decreased after starting a healthy eating plan, what is not yet clear is their experience of fatigue and how fatigue affects their motivation/ability to commit lifestyle changes.

CHAPTER 5: THE EXPERIENCE OF FATIGUE IN WOMEN WITH OBESITY: A QUALITATIVE STUDY

Two systematic style reviews and two surveys in this thesis have investigated the relationship between ID, fatigue and obesity in women who are obese using quantitative research methods. However qualitative research, which is often defined as "research beyond numbers" (Greenhalgh & Taylor, 1997), was required to enrich the available data on this subject by exploring women's experiences regarding fatigue.

The overall aim of this qualitative study was to increase depth of information available regarding the fatigue experience of women with obesity and how it influences their motivation and ability to commit to lifestyle change.

The objectives of the study were;

I. To determine experience of fatigue in a population of Slimming World female members.

II. To determine how fatigue influences their motivation and ability to commit to lifestyle change.

II. To determine how dietary changes affect their fatigue level.

III. To determine how weight loss affects their fatigue level.

IV. To explore their beliefs about how reducing fatigue levels may further improve their quality of life.

This chapter describes the methods used in the qualitative study and the findings which emerged from it.

5.1 METHOD

5.1.1 Ethical considerations

Ethical approval was obtained from University of Nottingham School of Biosciences research ethics committee (SBREC170109A) on 16/11/2017 (Appendix X).

All participants received a £15 Amazon voucher as an incentive for taking part.

5.1.2 Study design, sampling and data collection

The qualitative study compromised of two methodological phases as phase 1: focus group and phase 2: telephone interview. The following questions were developed and the same semi-structured interview guide was used for both phases. Additional follow-up questions were asked, as appropriate, with each interview.

Sample questions for semi-structured interview

- Have you experienced fatigue?
- If yes, how has fatigue affected you?

Probes: What has been the most difficult case related with fatigue that you have faced? What kind of issues arose?

- How do you feel fatigue influences your motivation/commitment to lifestyle changes?

Probes: What are the barriers to lifestyle changes related with fatigue? How do you address these?

- How does fatigue influence your ability to be active?

- Do you feel that your levels of fatigue have changed since you joined your Slimming World group?

Probes: What do you think about the reason for this decrease/increase? Do you think this change is related with diet? Do you think this change is related with weight loss?

- Do you feel that any changes in fatigue levels may improve your quality of life when you reach your target weight?

5.1.2.1 Focus group

Female members of Slimming World (from local groups and aged 19-49 years) were invited to take part in a focus group where a semi-structured interview process was used. In order to provide convenient access to University Park Campus, only the members of particular SW groups (Beeston, Chilwell and Nottingham) were invited to the study. The invitation was online via the SW Facebook group pages following communication between the Nutrition & Research team at SW Head Office and the local SW group facilitators.

Women who wished to take part in the focus group contacted the researcher and information sheets were sent to them via email. After obtaining consent, participants were invited to join the focus group held in a pre-booked room on campus. Although attendance of four women was anticipated, only two of those actually attended the focus group. The focus group was held with two facilitators at the University Park Campus, University of Nottingham. At the beginning of the session, the aim of the focus group was explained by the researcher and a

relaxed and comfortable conversation was created. The focus group was conducted in January 2018 and lasted 21 minutes. Tape-recording and note-taking were employed to keep an accurate record of the discussions and participants were informed about these processes.

5.1.2.2 Telephone interviews

Female members of SW were invited to take part in telephone interviews without considering their ages. The invitation was made by the local SW group facilitators and all telephone interviews were managed by a MSc Clinical Nutrition master student between February and March 2018.

The aim of the interview was explained by the student at the beginning of each telephone conversations and a semi-structured interview was carried out. All interviews were audio recorded by using a personal mobile phone. Data on age, ethnicity, use of medications, weight and height were gathered via emails after the telephone interviews. . Four women completed telephone interviews, with each telephone interview lasting between 6-8 minutes.

In terms of sample size, whilst it is calculated on the basis of the total population size and margin of error in qualitative studies (Campbell et al., 2010), Sandelowski (1995) described adequacy of sample size in qualitative research as "relative, a matter of judging a sample neither small nor large per se, but rather too small or too large for the intended purposes of sampling and for the intended qualitative product". Morgan (1996) suggested that data saturation should remain the underlying factor that influences the number of participants in qualitative research. In this study, interviews were conducted until new data did not reveal new themes or insights (Creswell, 2009). A clear pattern and data

saturation emerged with six participants in total from focus group and telephone interviews. Guest et al. (2006) consistently showed that the first six interviews might be enough to achieve theoretical saturation.

5.1.3 Data analysis

There are number of methods for undertaking qualitative data analysis including grounded theory; discourse, conversation, content and thematic analysis (Smith, & Firth, 2011). Thematic analysis, which is defined as the process of identifying patterns or themes within qualitative data to provide an informative description of the phenomenon (Smith, & Firth, 2011; Maguire & Delahunt, 2017), was employed for the study. A good thematic analysis interprets and makes sense of the data, rather than simply summarising it (Maguire & Delahunt, 2017). Although many different ways to approach thematic analysis are available (Alhojailan, 2012; Javadi & Zarea, 2016), the six-step framework by Braun & Clarke (2006) was followed in this study.

The framework (Braun & Clarke, 2006) included the following steps: 1) Becoming familiar with the data, 2) Generating initial codes, 3) Searching for the themes, 4) Reviewing themes 5) Defining themes and 6) Writing-up

5.1.3.1 Becoming familiar with the data

This step included listening to audios, reading transcripts and studying field notes. This stage helped the researcher to list key ideas including words/phrases that were frequently used by participants. Each audio recording was listened to at least twice and all recorded audio interviews were transcribed verbatim. The

transcripts were read at least twice to be able to be very familiar with the entire body of data.

5.1.3.2 Generating initial codes

In this phase, the data was organised in a meaningful and systematic way. “Open coding” methodology was employed and pre-set codes were not identified. The codes were developed and modified as it was worked through the coding process (Maguire & Delahunt, 2017).Open coding was undertaken by using a mixed method approach (both manually and data analytic software). First, hardcopies of the transcripts were worked through with pens and highlighters. Secondly, all Microsoft Word transcript files were loaded into NVivo12 (QSR International, Doncaster, Vic., Australia) software to create and organise nodes that are collection of references about a specific theme, case or relationship (QSR International, 2018)

5.1.3.3 Searching for themes

A theme represents significant or interesting patterned meaning about the research questions within the data (Maguire & Delahunt, 2017). According to Braun & Clarke (2006) there are no hard and fast rules in relation to what makes a theme. At this stage, the different codes were sorted into potential themes and subthemes using NVivo. Due to small sample size (n=6), this stage of identifying preliminary themes overlapped with the previous stage (generating initial codes).

5.1.3.4 Reviewing themes

At this stage, all the data related to each theme and subtheme was gathered together. The initial themes and subthemes were reviewed, modified and developed. Following points were considered at this stage; meaning of the themes, supporting the themes by the data, capacity of a theme (fitting too much into a theme), separating themes if the themes overlap, existing of subthemes (themes within themes) and existing of other themes within the data (Maguire & Delahunt, 2017).

5.1.3.5 Defining themes

This stage was aimed to identify the "essence" of meaning of each theme and as well as the themes overall (Braun & Clarke, 2006). Themes were defined and named. Clear and emphatic names were selected to provide the reader a perception of what the theme is saying.

5.1.3.6 Writing-up

Findings from the thematic analysis were reported in section 1.2.2 in this chapter.

5.2 RESULTS

5.2.1 Sample description

Six participants participated in the qualitative study (two in focus group and four in telephone interviews). All participants were women who were overweight or obese (BMI: 25≥kg/m^2). The mean BMI of women who participated the telephone interviews was 33.0 kg/m^2 (table 5.1)

Table 5.1 Characteristics of the participants who completed a telephone interview

	Age (years)	Race	Weight (kg)	Height (cm)	BMI (kg/m^2)
C*	34	WB	114.3	175.3	37.2
J*	47	WB	70.5	157.5	28.4
K*	28	WB	85.2	150.8	37.5
T*	53	WB	110.0	172.8	36.9
Mean	40.5	-	95.0	164.1	35.0

*Participants were coded based on their initials.

WB, white British

5.2.2 Findings

The main purpose of this qualitative study was to develop understanding of women's fatigue experience and to identify its effects on their motivation and ability to change their lifestyle.

Five broad themes emerged from the analysis: Experience of fatigue, effects of fatigue, overcoming fatigue, quality of life related fatigue and weight stigma (table 5.2).

Table 5.2 Themes and subthemes identified from analysis of transcribed interviews

Themes	**Subthemes**
Experience of fatigue	-
Effects of fatigue on women's life	Day-to-day activities Walking and physical activity Lifestyle changes
Ways to overcome fatigue	Joining Slimming World Healthy diet Weight loss Other factors
Quality of life related fatigue	-
Weight stigma	-

5.2.2.1 Experience of fatigue

Concerning experience of fatigue, a common answer amongst participants, when asked "Have you experienced fatigue?", was: "Yes, I have".

A participant said: "I was definitely more fatigued before I started Slimming World."

5.2.2.2 Effects of fatigue on women's life

A recurrent theme in the data was a sense amongst participants that fatigue affected many aspects of their lives including their daily activities, physical activities and motivations.

One participant described fatigue as: "It just meant that it took a lot longer to get things done really."

Similarly, a woman said: "I guess I've noticed it's harder to do things…."

Another participant said: "I know when I'm physically tired and fatigued, which I would put down to the fact that I am overweight because, you know, of the excess strain on my body that's, kind of, in some ways to me quite obvious that it's going to have an effect on you….."

Day-to-day activities

The majority of women that completed the telephone interview stated that fatigue affected their daily activities including their work.

"I have two young children as well, so I can find it difficult to interact with them, play with them, you know. Sometimes, because I do work full time as well, you know, afternoon naps are required……."

"It impacts on work as well, focus, if I focus, for focus and sleeping as well because you sleep too much…."

"Just really tired all the time, lethargic and just daily activities and things like that, you know, just general day-to-day things, you know, like exercise and things like that were…yes, feeling, sort of, like, sluggish…."

Walking and physical activity

A common view amongst the women that took part in the focus group was that fatigue had negative impacts on their physical activities including walking, walking up the stairs and exercise.

"…….. I just get more tired a lot more quickly and just doing normal things, like, walking up the stairs, those kinds of things……. I've noticed I get tired just, sort of, walking a lot now if I'm walking a long distance and although it's not a cardio thing, like, running up the stairs or anything…."

"I would agree with that, like, just going upstairs and stuff there's a noticeable difference to now as to how…even back just to December, in September when I was running up the stairs to my room I'd be really tired and now I've lost a little bit of weight it's better, but it's not what it used to be. Yes, so it's quite easy to get tired."

Both women mentioned that more weight was related with more tiredness despite the same physical activity. Hence, those women preferred to avoid doing exercise or spent more time for a particular activity to prevent fatigue.

Talking about this issue a participant said: "……. like the more fatigued you are the more you stop doing and then it just, kind of, spirals downwards. Yes, it's hard to, sort of, force yourself to do those things initially……."

A participant said: “I think also it’s almost just the fear knowing that you’re going to be fatigued after doing something. So, I put off going upstairs because I know I’m going to be out of breath at the top and I almost don’t even want to face the fact that I’m going to be tired once I get to my room because I feel like I’m failing myself with the fact that I can’t do a simple thing, like, go upstairs.”

Lifestyle changes

In terms of effects of fatigue on motivation and ability to commit to lifestyle change, the majority of women considered that fatigue negatively influenced their motivation to change their lifestyle such as going to gym.

A participant said: “It makes it harder, yes, definitely because I’m a relatively physical person. I spend a lot of time on my feet, just because I’m either doing singing practice or I’m working at the leisure centre or I’m just doing chores at home. But, then having the motivation to go to the gym or go swimming is…yes, it’s more difficult.”

Another participant said: “……. Even though I know…I keep saying to myself, oh I’ll start going to the gym more often, and I know if I went every day for a week I’d probably be fine, and the fatigue will go down a bit, but, yes, it’s just…it just seems like so much more of a massive undertaking now, yes.”

Views of women that took part in the telephone interview were similar.

“…… if you’re tired you don’t want to do anything, you know. If you’re feeling sluggish and suffering from fatigue your motivation levels are a lot reduce and, yes, you just don’t feel like engaging in much, especially anything that’s particularly energetic……. with the lack of motivation to do stuff I’m,

kind of, not necessarily wanting to participate in activities that I feel like might be too tiring…"

"In terms of fatigue how it affects me, it effects my mood, my ability to be motivated…… It's easy to deflect it, put it off for another day."

5.2.2.3 Ways to overcome fatigue

In terms of the methods that were used to overcome fatigue, a variety of perspectives were expressed.

Joining Slimming World

The majority of women reported that joining Slimming World helped them to overcome fatigue over and above losing weight. The quotes below illustrate this:

"I think joining Slimming World is, kind of, a massive thing for me……. So, I think if I can get a bit…regardless of where I am in my weight I always think if I can lose half a stone then I'll join the gym and you'll notice, sort of, a difference and it will encourage you to start doing more……"

"I'd agree with that. My Slimming World definitely helps."

"……. I think, I suppose, I'm feeling a bit more positive because, obviously, I'm attempting to lose weight, so joining the group and feeling that bit more motivation…." (J-TI)

"……. it's really helped me basically, it's saved me, so…yes, yes."

Healthy diet

Views related to diet were particularly prominent in the data. Majority of the women stated that the SW diet and healthy eating contributed to reduce their level of fatigue.

“The diet has actually made a really big difference…….”

“Yes, it definitely, like, if I’m eating more healthy, I notice that I’ve got more energy. If you eat junk food it makes you very, sort of, lethargic and sleepy anyway. Again, if you’re eating portions that are too big then you, kind of, get this food coma afterwards……”

“I think it’s related with diet because I used to eat an awful lot of bread as well and potatoes and pasta and I find that brings me…that drains you. ….. Yes, I do believe that it has helped to improve because I’m eating so much better. I’m not eating rubbish. I’m not eating the things that used to make me feel quite blah.”

“Because, I’m like cooking from scratch and know what goes in the meals, it’s more healthy eating, it’s increased your metabolism and wanting to do things. I’ve got more energy, like, for the grandkids and just going out more and, you know, wanting to take part in exercise and things like that, yes…. I definitely think the diet has helped, you know, the healthy lifestyle changes have helped me, you know, with my mood and wanting to do more exercise, things like that. And, also you get a bit of buzz for it as well, you know, like, when you’re planning and preparing meals, that’s a massive part in it and it does keep you focused……”

Perspective of a participant that was different from the others:

"…… I did a week of SP last week and when I cut out the carbohydrates completely there were…my level of fatigue seemed to fluctuate more rather than being at a constant level. When I'm eating pasta and all of that…if you eat whole grain pasta that's a slow burner, you're going to have that energy for a while. Obviously, with SP when you're cutting out that carbohydrate you don't have that, so I'd be really, really energised for an hour after eating and then I'd plummet, and I'd be really tired. Then the next time I'd eat it would go back up again, so it was almost harder to control my level of fatigue because I didn't know when I was going to be hit with it, if that makes sense."

Weight loss

In terms of weight loss, a common view amongst participants was that when they start losing weight they feel a decrease in their levels of fatigue. The quotes below show this:

"Yes, I've got more energy. I'm obviously carrying a lot less weight around with me, but I don't think…when you've got weight on you it goes on so gradually, I don't think you realise how fatiguing it is to carry all that extra weight around with you. So, I've lost about two and a half stone so far and, yes, I just feel a lot better about myself as well…. Yes, I've got more energy to play with my little boy…. But, losing weight actually makes you feel quite energised as well and I hadn't realised that that would be the case without the exercise element really."

"……. how much extra weight you're carrying around and you imagine if you were actually physically carrying it in your hands how tired it would make you and you think, oh, I'm walking around with all that extra weight on me and

having to carry it. ……. The more weight you can get off then the easier everything becomes really."

"…… but there has been a correlation with when I've lost weight I haven't been as tired…."

"…… With just a little bit of general knowledge of biology if you've got less weight to carry you're generally going to be a little bit more able to do things, so I'm hoping they will come hand in hand, that as I lose weight I will feel physically fitter therefore won't feel as fatigued and tired. That's what I'm definitely hoping for because then I can actually play with my kids."

Other factors

The quotes below shows the other methods that were applied by the participants to overcome fatigue.

"Just trying to sleep when I could and, trying to get things done earlier in the day when I've still got more energy…….. and just same old things really, you know, sitting down and making a list and trying to get through it, if that makes sense."

"…… Then also there are the tips, like, making sure that, like, if you've just gone for a walk or something or you've come back home always walk in, have a glass of water because that will just refresh you a bit more……."

5.2.2.4 Quality of life related fatigue

The majority of the participants emphasised that improvement in fatigue levels may enhance their quality of life, when they reach their target weight as these

quotations illustrate:. "Yes, so I think I assume that once I get down to my target weight I'll be less fatigued. I mean, obviously, there are a lot of other issues, but I feel for me my weight is probably the biggest thing that's, sort of, keeping me fatigued constantly…… Then also I think things like concentration and those kinds of things will improve as well, so I'll be able to focus more on my work and things like that. If you're being more active…before I put the weight on I was much more active, and I did more, and I've definitely found it made a big difference in my work as well, like, I could think a lot more clearly."

"Yes, it will definitely improve my quality of life….. As a performer you need to…I need to have energy for any performance I do, and singing is so physical that if I don't have… If I'm carrying three and a half stone while I'm doing that, like, that's never going to happen as well as it could do if I wasn't holding it. So, I'm looking forward to getting to target and being able to do what I love better…… yes, it would definitely improve my life as to not have quite as high a level of fatigue."

"I hope so. I've seen improvements already, you know, I've gone back to the gym a couple of times. I've started badminton with my youngest son, just once a week, whereas before I was doing absolutely nothing….. So, even though I still have to force myself, because there is still the fatigue…. I'm more motivated to go and do something rather than collapsing and eating all night once I get in from work."

Whilst the majority of the women agreed that their quality of life will improve when they reach their target weight, one participant from the telephone interview said:

"I don't really know, to be honest. I can't see it changing from where I am now, just, like…but who knows. Do you know what I mean? It's a bit hard to see that far all the way down the line. I just feel good in myself, confidence wise..."

5.2.2.5 Weight Stigma

Although the study was not designed to identify weight stigma experiences of the participants, half of the participants mentioned their experiences and feelings about weight stigma related with fatigue.

"Yes, because I look at going to the gym and then I'm like, I'm going to be on a treadmill next to someone who can run a marathon and I'm there and I'm fatigued after, what, five minutes. Yes, knowing or just thinking of the situations that you could be put in or almost how you're going to be viewed because you're fatigued that, kind of, puts you off doing that…."

"Yes, you don't want to get puffed out within ten minutes of being there [chuckle]. I just feel like other people will judge me, so, yes, that's a big issue…."

"Yes, and it's not until you notice those changes yourself and then people tell you and little changes you notice, like, at work I used to run up the stairs and I'd be absolutely breathless, you know, and people would say to me, you know, 'are you okay?' It's quite embarrassing……"

5.3 SUMMARY AND DISCUSSION OF KEY FINDINGS

Findings from the qualitative study (n=6) showed that all participants experienced fatigue that influenced many aspects of their lives including their daily activities, physical activities and motivations. This finding broadly supports the quantitative work of other studies (Maffiuletti *et al.*, 2007; Anandacoomarasamy *et al*., 2009; Impellizzer *et al.*, 2013) in this area linking with the high prevalence of fatigue with obesity. There are also similarities between the experiences expressed by the women in this study and those described by African American women who are obese in a qualitative study (Befort *et al*., 2008). In this study (Befort *et al*., 2008), participants in focus groups at a community health clinic complained about low energy, feeling tired, and having difficulty breathing with only minimal exertion. They mentioned about their desires to lose weight because they want to improve their daily functioning. Similar to the findings from our study, many women wanted to lose weight to have more energy to play with their children or grandchildren. One of the participants expressed her perspective as "I think a healthy size for me would mean, it wouldn't really be a size as much as it would be a freedom to just do whatever I want to do. Just anything because so much, so many things I just don't do because I'm tired".

In terms of physical activities, women indicated that they found it difficult to do exercise, walking up the stairs and walking. Since fatigue impacted their motivation and ability to commit to lifestyle change, they reported their unwillingness to participate in activities such as going to gym or swimming. In another qualitative study (Thomas *et al*., 2008) explored attitudes towards

dieting, physical exercise and weight loss solutions in people with obesity. Findings from open-ended interviews indicated that the majority of participants found exercising difficult due to their weight and physical health problems. Also similar to the findings from our study, participants also mentioned that they were emotionally humiliated, embarrassed or daunted when they attempted to exercise alone.

Women in this study believed that joining SW helped them to overcome fatigue over and above losing weight. In a synthesis of qualitative research on overweight and obese people's views and experiences of weight management (Garip & Yardley, 2011), improvements in physical and psychosocial functioning that were not necessarily linked to weight loss were reported as positive outcomes of participating in a weight management programme. In our study, other factors that were reported as overcoming fatigue were losing weight and healthy eating. This finding agrees with the findings of other studies in this thesis (two systematic style reviews and two quantitative studies). Two systematic style reviews in this thesis showed that weight loss is effective to both improve iron levels and reduce inflammation in women who are obese. Similarly, two quantitative studies indicated that weight loss is associated with reducing level of fatigue. In a qualitative study by Jones *et al.* (2007) semi-structured interviews (n=24) explored the views of participants who were obese and attending dietetic clinics with the dietetic service and the treatment outcomes, in terms of lifestyle changed.

The majority of participants reported that they felt healthier and lighter with having more energy and found it easier to do many things.

In terms of healthy eating, some participants in this study claimed that "junk food" and high carbohydrate foods mentioned as "bread as well and potatoes and pasta" in the interviews increased their level of fatigue. The results of the second systematic style review in this thesis suggested that decreasing consumption of energy-dense foods is related with reducing inflammation in women who are obese. In addition, Study 1 in this thesis showed that participants who adhere to a "CHO and high fat" dietary pattern were more likely to have fatigue and those who followed a "vegetables" dietary pattern were less likely to have fatigue. All these findings might be explained by the relationship between antioxidants, dietary fibre, oxidative stress, inflammation and fatigue.

In this study, the majority of women expressed that their quality of life would be improved when they reach their target weight since their fatigue level would reduce.

Several limitations to this qualitative study need to be acknowledged. First, the small size of the focus group may have limited the range of fatigue experiences. Secondly, although motivation and mood might be significant factors that are affected by levels of fatigue, these factors were not presented in the themes. Because the research question related to motivation addressed lifestyle changes, participants might tend to express their fatigue experience related to motivation as part of their perspectives of lifestyle changes.

In conclusion, the majority of women in this study mentioned that fatigue had negative effects on their daily activities, desire to walk, do exercise and make lifestyle changes. They believed that their fatigue level decreased/would decrease by joining SW, eating healthily and losing weight, and reduced fatigue

would help them to improve their quality of life and motivation/ability to commit lifestyle changes. These results corroborate the findings of a great deal of the previous work (systematic style reviews and quantitative studies) in this thesis. Also, this study further supports the importance of multi-component weight management interventions that include supporting of healthy eating and more physical activity, and behavioural change techniques on not only weight loss but also psychosocial health. Hence, people's motivation to weight management efforts should be increased by highlighting the benefits of weight loss concerning the physical and psychosocial health.

CHAPTER 6: GENERAL DISCUSSION

This chapter discusses the findings which emerged from two systematic style reviews, two quantitative studies and one qualitative study that make up this thesis. It presents comparison of the findings from the UK (study 1) and Turkey (study 2).

6.1 DISCUSSION OF KEY FINDINGS

6.1.1 Low iron stores, ID or IDA status in women who are obese

Study 1 showed that approximately 40% of women experienced low iron stores, ID or IDA up to of the time they joined SW. However, only 9.6% of those experienced low iron stores, ID or IDA in the last three months after becoming a member of SW. It is encouraging to compare this figure with that reported in NDNS Report of Years 7 and 8 (combined) of the Rolling Programme (2014/2015-2015/2016) which indicated that 7% of women aged 19-49 years (defined by NDNS as "childbearing age") had low dietary iron intakes below LRNI and 8% of adult women (19 to 64 years) had IDA (Hb<120g/l) and 12% of those had low iron stores (ferritin<15µg/L) (Roberts *et al*., 2018). Although these findings from study 1 are consistent with the NDNS data, the prevalence of women reported that they had low iron stores, ID or IDA until consulting a dietitian in Turkey was higher (44.4% at T0 and 19.0% at T1). One of the strengths of this study is that these findings are supported with blood test results which indicated that the prevalence of ID was 23.7% and low iron stores was 28.9% in Turkish women who are obese.

6.1.2 Changes in body weight, fatigue and physical activity levels from baseline to follow-up

This thesis found that there was a decrease in BMI and an increase in PA levels from baseline to follow-up in both quantitative studies. These results are in line with those of previous studies. In a single-blind randomised trial (Goodpaster *et al*., 2010), the effects of one-year intensive lifestyle intervention consisting of diet and physical activity on weight were analysed. A statistically significant decrease in BMI was found in both groups (group 1: diet and physical activity for the entire 12 months, group 2: identical dietary intervention but delayed physical activity for 6 months). Regarding PA, although the initial-activity group had significantly increased their PA over the 12-months period (from 7048 steps to 8475 steps per day, $p<0.001$), the delayed-activity group did not significantly increase their PA in the first 6 months. However, between 6 and 12 months, the delayed- activity group significantly increased their physical activity (from 7047 steps to 7991 steps per day, $p=0.01$). In another randomised controlled trial (Fuller *et al*., 2014), the same physical activity assessment method (IPAQ-short form) was used to evaluate PA over the period of weight loss (12 months) and follow-up (24 months) for standard care, as defined by national guidelines and for a commercial programme (Weight Watchers). Similar to our findings, there was a statistically significant increase in PA from baseline to 12 months and 24 months in both groups.

In terms of fatigue level, quantitative studies and qualitative study in this thesis indicated that weight management is key in terms of reducing fatigue level which might be related with ID, inflammation and/or obesity in women who are

obese. Both quantitative studies suggested that there was an improvement in fatigue level from baseline to follow-up and our results are in agreement with a six month intervention of PA support treatment paired with cognitive-behavioural nutrition for weight loss which showed significant improvements in fatigue (F=133.35, p<.001, multivariate partial eta squared=.450) (Annesi *et al*., 2015). Consistent with these findings, the qualitative study in this thesis found that participants reported weight loss as an important factor to overcome fatigue.

6.1.3 Associations between body weight, iron status, fatigue and levels of PA in women who are obese

The present study indicated that although the mean BMI of women in the UK study was higher than Turkish women at baseline (35.0 vs. 32.4 kg/m^2), the GFIs at baseline were similar in both populations (29.2 vs. 28.1). However, the percentage of decrease in GFI from T0 to T1 was greater in women in the UK than Turkish women (25.3% vs. 1.4%). In line with the findings related to fatigue level, the percentage of increase in total PA from T0 to T1 was greater in women in the UK than Turkish women (124.7% vs. 58.1%). It seems possible that these results are due to bi-directional relationship between fatigue and PA. Whilst women who feel less fatigued are more likely to be motivated to do physical activity, women who do more physical activity are less likely to feel fatigued. It is in accord with the qualitative study in this thesis indicating that fatigue impacted women's PA, motivation and ability to commit to lifestyle change negatively. Similarly, Annesi *et al*., (2015) found that increased physical activity predicted reduced fatigue (β = –.192, SE = .029, p = 0.014).

The most effective method of weight loss which reduces obesity related health risks is still not clear. In study 1, PA level at T2 was significantly and positively correlated with weight loss. In a systematic review (Johns *et al*., 2014), randomised controlled trials of combined behavioural weight management programs and diet-only or physical activity-only programs were compared. Analysis of six studies indicated that from baseline or at 3 to 6 months, no significant difference in weight loss was found between the combined behavioural weight management programs and diet-only programme. However, in the long term (at 12 months), it was reported that the combined behavioural weight management programs provided a significantly greater weight loss. In comparison of the combined behavioural weight management programs with physical activity-only, analysis of five studies showed that physical activity-only programs were less effective in both short (3 to 6 months) and long term (12 to 18 months).

In terms of the relationship between weight loss and fatigue, the findings from the hierarchical multiple regression in the study 1 suggested that after controlling the effect of PA at T0, T1 and T2, baseline BMI and decrease in fatigue level explained only 9.5% of the variance in weight loss. Although, the ability of fatigue level to predict weight loss was not very strong, a significant positive correlation between decrease in GFI and weight loss was found. The qualitative study in this thesis also showed that more weight was related to greater level of fatigue in women who are obese. Bryant *et al.* (2008) examined the relationship between vital exhaustion and BMI in white and African-American adults (n= 13,727). The study showed that there was a significant positive correlation between BMI and vital exhaustion among both white and African-American

adult and baseline vital exhaustion predicted future excess weight gain in white men and women. The results reported here may support the hypothesis of impaired iron status which leads to fatigue in people who are obese. Study 2 in this thesis showed that Hb, haematocrit and TIBC levels were inversely associated with both BMI and fatigue level in Turkish women who are obese. These results also mirror those of the systematic style reviews in this thesis which reported that weight loss and exercise are effective to both improve iron levels and reduce inflammation in women who are obese. This combination of findings provides some support for the importance of considering ID or low iron stores in women who are obese before starting a weight management programme.

6.1.4 Food frequency and iron-related dietary patterns

Analysis of FFQ showed that "milk added to drinks" and "coffee" were the two most frequently consumed food groupings of women in the UK at all the time periods (T0, T1 and T2) and "black tea" was the most consumed food grouping of Turkish women both at T0 and T1. Both "coffee" and "black tea" are known to be potent inhibitors of iron absorption. Reported mean frequency of consumption of black tea per week was 3.77±8.09 in women in the UK and 15.9±9.6 in Turkish women at baseline. Tea consumption was reported to be 1.9 kg per capita per year for the UK and 2.9 kg per capita per year for Turkey by Food and Agriculture Organization of the United Nations (FAO), (2013). Despite the high consumption of tea, consumption of meat as a good source of haem iron was considerably lower in Turkey compared to the UK. Meat consumption (including bovine meat, mutton & goat meat, pig meat, poultry meat) was reported to be 80.0 kg per capita per year for the UK and 35.1 kg per

capita per year for Turkey (FAO, 2013). Higher consumption of tea and lower consumption of meat products might explain the high prevalence of ID in women of reproductive age group in Turkey.

Since study 1 was limited by the lack of iron-related blood parameters and study 2 was limited by the relatively small sample size, it was not possible to explore the relationship between iron status and tea/coffee related dietary pattern. However, study 1 investigated the association of presence of fatigue with "milk and coffee" dietary pattern at T0 and "milk, tea and coffee" dietary pattern at T1. No differences were found in terms of the relationship between these two dietary patterns and presence of fatigue. Beck *et al.* (2013) investigated associations between dietary patterns and suboptimal iron status (serum ferritin <20 μg/l) in premenopausal women aged between 18 and 44 years (n=375). The same iron FFQ used and seven dietary patterns as "refined carbohydrate and fat", "Asian", "healthy snacks", "meat and vegetable", "high tea and coffee" "bread and crackers" and "milk and yoghurt" were identified. Findings from logistic regression suggested that following ''milk and yoghurt'' and ''high tea and coffee'' dietary patterns were associated with an increased risk of low iron stores. However, there was an inverse correlation between "meat and vegetable" dietary pattern and the risk of suboptimal iron status.

Study 1 found that participants who adhere to the "CHO and high fat" dietary pattern were more likely to have fatigue at T0 and those who adhere "vegetables" dietary pattern were less likely to have fatigue at T1. These results match those observed in earlier studies. In a randomised exercise study (Guest *et al.*, 2013), the relationship between dietary intake and fatigue were

determined in breast cancer survivors (n=42). Results from 3-day diet record and Functional Assessment of Cancer Therapy for fatigue showed that there was a positive correlation between fatigue and percentage of energy intake from fat per day ($r = 0.31$, $p < 0.05$) and a negative correlation between fatigue and dietary fibre intake (g) per day ($r = -0.38$, $p < 0.05$). In contrast to our findings, however, fatigue was inversely related to dietary carbohydrate intake (g) per day ($r = 0.31$, $p < 0.05$). In the qualitative study in this thesis, the findings into the relationship between CHO intake and fatigue were also inconsistent and contradictory. Whilst a woman claimed that eating less CHO helped to improve her level of fatigue, another woman asserted that her level of fatigue seemed to fluctuate more rather than being at a constant level when she reduced CHO intake.

Regarding associations of dietary patterns with BMI, study 1 found that there was a positive association of adherence to "CHO and high fat" and "meat and eggs" dietary patterns with a higher BMI at T0 and "healthy CHO and eggs" dietary pattern with a higher BMI at T1. However, "milk, tea and coffee" dietary pattern was related to lower BMI at T1. These results seem to be consistent with Cho *et al.* (2003) who investigated the relationship between breakfast type, energy intake and BMI. Results from the Third National Health and Nutrition Examination Survey, a large, population-based study conducted in the United States from 1988 to 1994, indicated that subjects who ate meat and eggs for breakfast had the highest daily energy intake and higher BMI compared to other breakfast groups including skippers, ready-to-eat cereals, fruits and vegetables, and breads.

6.1.5 Dietary habits

Study 1 found that the percentages of participants who reported they always had breakfast increased significantly from T0 to T1. This habit might be one of the underlying factors for successful weight loss and management. Wyatt *et al.* (2002) examined breakfast consumption in subjects maintaining a weight loss (n=2959) by a cross-sectional study. Subjects, (members of the National Weight Control Registry in the United States), on average, lost 32 kg and maintained it for 6 years. Whilst 78% of the participants reported that they were regularly eating breakfast every day, only 4% of those reported never eating breakfast. It was concluded that eating breakfast was one of the habits associated with weight loss maintenance. In contrast, study 2 found that the percentages of participants who reported they always had breakfast decreased from T0 to T1. These results, however, need to be interpreted with caution due to small sample size.

In both study 1 and 2, the majority of participants reported that they usually have a drink up to 1 hour before, with, or up to 1 hour after their breakfast, lunch and evening meal at all the time points. Tea was one of the most consumed drinks consumed before, after or with meals in both the UK and Turkish populations. In terms of types of food consumed during main meals (breakfast, lunch and dinner), reported consumption of bread, toast or starchy food reduced and vegetables increased from T0 to T1 in both populations. Changes in the types of food and drinks consumed between meals and after the evening meal were also similar between the UK and Turkey. Consumption of junk food, including biscuits or cakes, potato crisps, chocolate or sweets decreased, and fruit and vegetable intake, increased from T0 to T1. Changes in the types of food and

drinks as part of the weight management interventions might be related with changes in fatigue levels. The second systematic style review in this thesis suggested that fibre- and antioxidant-rich foods such as fruits and vegetables are associated with reduced inflammation by reducing oxidative stress.

6.2 LIMITATIONS

The overall aim of this PhD was to investigate the potential links between obesity and ID, particularly in women of child-bearing age. Although the study successfully demonstrated that fatigue, one of the most common sign of ID, was closely related with higher BMI and there was a correlation between Hb, haematocrit, TIBC, BMI and fatigue, it has certain limitations in terms of exploring impaired iron status in women who are obese. Firstly, the studies did not assess participants' serum hepcidin level which is influenced by low-grade inflammation due to excess adiposity and reduces iron absorption. Secondly, in spite of the availability of iron related parameters at baseline in study 2, it was unfortunate that the study did not include the blood test results at follow-up. Also, the small sample size in study 2 limits the generalisability of its findings.

Although one strength of study 1 was collecting the data for T0 and T1 using the same survey at the same time, both study 1 and 2 were limited by the absence of large number of participants at follow-ups (T2 in study 1 and T1 in study 2). One of the reasons for this might be related with a large number of questions in the survey and no longer attending the SW groups or dietetic consultations. Shayna *et al.* (2017) investigated specific barriers and facilitators for engagement and weight loss in a workplace weight management program by a qualitative study (n=28). It was suggested that numerous barriers influenced

participants' engagements in workplace weight management programs and the coaches had a strong impact on their engagement in the program. Focussing on changing needs, increasing frequency and variety of contact by coaches and tailored educational materials were suggested by participants. Another reason for the high percentage of participants who were not included at follow-up (T2) in the Study 1 was the absence of a strong question to match the data. Although SW membership numbers and e-mail addresses were used to match data, participants' date of birth could have been asked again to support data matching and gain greater sample size at follow-up.

The qualitative study in this thesis provides a novel approach to identifying fatigue experience of women who are obese by a focus group and telephone interviews. One limitation of the qualitative study was that the focus group were carried out with only two participants. However, in the second phase of the study, four more participants were interviewed, and saturation was achieved. Draper and Swift (2010) suggested that large or statistically representative samples are not required for most qualitative research because the aim is theoretical generalisation.

6.3 RECOMMENDATIONS FOR FUTURE RESEARCH

The first systematic style review in this thesis found that additional dietary iron in very low energy diet, high-protein diet and weight loss, breakfast cereals and exercise training were effective on improving some iron related parameters in women who are overweight or obese. The second systematic style review suggested that aerobic exercise, higher protein, brown rice, histidine supplementation, and tomato juice were found to be effective to reduce

inflammation over and above weight loss. Study 2 supported the evidence that women with obesity have low-grade inflammation by providing an inverse correlation between BMI and TIBC and a positive correlation between ferritin and BMI. Both Study 1 and 2 showed that women's fatigue level decreased with weight loss and increased PA. Similarly, the qualitative study in this thesis revealed that women believed that their level of fatigue decreased, and quality of life improved when they reached their target weight. Although these studies and literature provide reasonably consistent evidence of an association between obesity and inflammation and its effect on ID, to date, no previous study aimed to improve iron status of women who are obese by reducing obesity related inflammation. Therefore, what is now needed is an intervention study that investigates if a strategy aiming to reduce inflammation could improve iron status in obese iron deficient women of reproductive age, by reducing hepcidin concentration, and removing the basolateral inhibition of iron absorption. Further studies regarding the role of histidine supplementation and tomato juice on improving iron status in women who are obese would be worthwhile, because these two intervention studies had positive effects on inflammation in obesity without providing a weight loss. To understand the association between ID and low-grade inflammation due to excess adiposity clearly, hepcidin levels should be measured in future work.

Although, the majority of previous studies have focused on adipose tissue as the source of obesity-associated inflammation, more recent studies have concentrated on gut bacteria. Ding *et al.* (2010) hypothesised that a high-fat western diet and gut bacteria interact to promote intestinal inflammation, which contributes to the progression of obesity and insulin resistance. In this study,

conventionally raised specific-pathogen free and germ-free mice were compared in terms of intestinal inflammation after giving a high-fat or low-fat diet for 2-16 weeks. It was reported that a high-fat diet induced ileal TNF-α mRNA in conventionally raised specific-pathogen free but not germ-free mice. This increase preceded obesity and weight gain. A strong and significant association between these pro-inflammatory changes and the development of obesity and insulin resistance were reported. Therefore, it was suggested that interventions which limit intestinal inflammation promoted by a high-fat diet and bacteria may be beneficial in combating risk of obesity and insulin resistance.

Bacterial products caused by metabolic disease-associated processes bring on a reduction in tight junction expression. Hence, whole bacteria translocation to the circulation increases because of enhanced intestinal permeability. As a result, an immune response, inflammation, and immune cell infiltration of liver and adipose tissue are triggered (Carvalho & Abdalla Saad, 2013). Insulin resistance occurs in various tissues by diverse mechanisms and food intake deregulation in the hypothalamus promoted by the insulin and leptin resistance (Carvalho & Abdalla Saad, 2013). Modulation of gut microbiota may enhance the intestinal barrier integrity, reduce circulating antigen load, and ultimately ameliorate the inflammation and metabolic phenotypes (Xiao *et al*., 2014). One of the most effective interventions to modulate gut microbiota may be the use of probiotics. Probiotics are defined as live microorganisms that confer unspecified health benefits to the host and prebiotics which are non-digestible food ingredients such as certain fibres (Caselli *et al*., 2013). Probiotic treatments are capable of modulating the gut microbiota profile, their products and metabolites. This contributes to a shift in the bacterial community prevalence. As a result of this,

tight junction expression and function increase, intestinal permeability and circulation levels of bacterial community reduce, and inflammatory status in insulin-sensitive tissues are reduced (Carvalho & Abdalla Saad, 2013).

Given the reasons above, a randomised controlled trial to identify effects of synbiotic supplementation, combining probiotics and prebiotics, on improving iron status in women with obesity and ID is recommended.

6.4 IMPLICATIONS FOR PUBLIC HEALTH AND DIETETIC PRACTICE

The most obvious finding to emerge from all studies in this thesis (systematic style reviews, quantitative studies and qualitative study) was that weight loss, reducing energy-dense foods and increasing foods with a high fibre and antioxidant content, such as fruits and vegetables, might be directly related with improving iron status, quality of life, ability to commit to lifestyle change and reduction of inflammation and fatigue in women with obesity. Given the most effective strategy for weight loss is considered to be a combination of diet, PA and behavioural changes, strategies to investigate underlying factors of ID or fatigue and to combat them will increase women's PA level and improve behavioural changes. In terms of dietary advice, personalised nutrition could be more effective than generalised dietary guidelines and "one-fit-for-all" dietary approaches to gain better understanding on underlying factors of ID or fatigue and provide more tailored interventions.

There are several possible explanations for the high rate of fatigue in women who are obese but one of the most common causes might be ID which is highly prevalent in women with obesity. Therefore, women with obesity who wish to

lose weight should have their iron status checked before starting an intervention. Dietary interventions should be designed to tackle ID and suggestions to improve iron related dietary habits such as timing and frequency of tea consumption should be given to women with ID. In Turkey, people who apply to a Nutrition and Dietetic clinic with the intent of losing weight are advised to consult a doctor to be checked their blood test results including fasting blood glucose, thyroid stimulating hormone and iron status (Ministry of Health of Turkey, 2017).

Co-existence of undernutrition and obesity presents not only at individual level, but also at population level. Therefore, governments of countries like Turkey which faces double burden of malnutrition (high prevalence of ID and obesity) should consider both undernutrition and overnutrition while planning public health policies to combat obesity.

6.5 CONCLUSION

The main goal of the current thesis was to investigate the potential links between obesity and ID, particularly in women of child-bearing age. This thesis provided a deeper insight into this subject with a systematic style review assessing the effectiveness of interventions designed to address ID in women of childbearing age who are either overweight or obese, another systematic style review exploring the effectiveness of interventions designed to reduce inflammation in adult women who are either overweight or obese, an online survey identifying any links between BMI, fatigue level, dietary patterns and dietary factors affecting the bioavailability of iron and PA levels in women of child-bearing age joining SW, a paper-based survey identifying any associations between BMI, iron status, fatigue level, dietary factors affecting the bioavailability of iron and PA level in Turkish women of child-bearing age and a qualitative study increasing depth of information available regarding fatigue experience of women with obesity and how it influences their motivation and ability to commit to lifestyle change.

Overall, this thesis confirms previous findings and contributes additional evidence that suggests that weight management has a positive impact on reducing inflammation, improving iron status, fatigue level, quality of life and motivation/ability to make lifestyle changes in women of reproductive age who are obese. Although reduced body fat as a result of weight loss might be a reason for all these positive effects, increased PA and consumption of fibre/antioxidant rich foods, and decreased consumption of energy-dense foods which are related with increased oxidative stress might be other factors contributing those positive

effects. Intervention studies are required to establish the effects of foods or supplements that have anti-inflammatory properties independently from weight loss, such as tomato juice, histidine and probiotic/prebiotic or synbiotic supplementations, on improving iron status in women who are obese by reducing inflammation.

REFERENCES

Abbaspour, N., Hurrell, R., & Kelishadi, R. (2014). Review on iron and its importance for human health. *Journal of Research in Medical Sciences: The Official Journal of Isfahan University of Medical Sciences*, *19*(2), 164.

Acaster, S., Dickerhoof, R., DeBusk, K., Bernard, K., Strauss, W., & Allen, L. F. (2015). Qualitative and quantitative validation of the FACIT-fatigue scale in iron deficiency anemia. *Health and Quality of Life Outcomes, 13*(1), 60.

Adolfsson, B., Carlson, A., Undén, A. L., & Rössner, S. (2002). Treating obesity: A qualitative evaluation of a lifestyle intervention for weight reduction. *Health Education Journal, 61*(3), 244-258.

Ahmad Fuzi, S. F., Koller, D., Bruggraber, S., Pereira, D. I., Dainty, J. R., & Mushtaq, S. (2017). A 1-h time interval between a meal containing iron and consumption of tea attenuates the inhibitory effects on iron absorption: a controlled trial in a cohort of healthy UK women using a stable iron isotope. *American Journal of Clinical Nutrition*, *106*(6), 1413-1421.

Alhojailan, M. I. (2012). Thematic analysis: A critical review of its process and evaluation. *West East Journal of Social Sciences*, *1*(1), 39-47.

Ainsworth, B. E., Haskell, W. L., Whitt, M. C., Irwin, M. L., Swartz, A. M., Strath, S. J., O Brien, W. L., Bassett, D. R., Schmitz, K. H., Emplaincourt, P. O. & Jacobs, D. R. (2000). Compendium of physical activities: an update of activity codes and MET intensities. *Medicine and science in sports and exercise*, *32*(9; SUPP/1), 498-504.

Alves, N. E. G., Enes, B. N., Martino, H. S. D., Alfenas, R. D. C. G., & Ribeiro, S. M. R. (2014). Meal replacement based on Human Ration modulates metabolic risk factors during body weight loss: a randomized controlled trial. *European Journal of Nutrition, 53*(3), 939-950.

Anandacoomarasamy, A., Caterson, I. D., Leibman, S., Smith, G. S., Sambrook, P. N., Fransen, M., & March, L. M. (2009). Influence of BMI on health-related quality of life: Comparison between an obese adult cohort and age-matched population norms. *Obesity, 17*(11), 2114-2118.

Anderson, R. A., Polansky, M. M., Bryden, N. A., & Canary, J. J. (1991). Supplemental-chromium effects on glucose, insulin, glucagon, and urinary chromium losses in subjects consuming controlled low-chromium diets. *American Journal of Clinical Nutrition*, *54*(5), 909-916.

Annesi, J. J., Johnson, P. H., & Porter, K. J. (2015). Bi-directional relationship between self-regulation and improved eating: temporal associations with exercise, reduced fatigue, and weight loss. *Journal of Psychology, 149*(6), 535-553.

Armah, S. M., Boy, E., Chen, D., Candal, P., & Reddy, M. B. (2015). Regular Consumption of a High-Phytate Diet Reduces the Inhibitory Effect of Phytate on Nonheme-Iron Absorption in Women with Suboptimal Iron Stores, 2. *Journal of Nutrition*, *145*(8), 1735-1739.

Armstrong, M. J., Mottershead, T. A., Ronksley, P. E., Sigal, R. J., Campbell, T. S., & Hemmelgarn, B. R. (2011). Motivational interviewing to improve

weight loss in overweight and/or obese patients: a systematic review and meta-analysis of randomized controlled trials. *Obesity Reviews*, *12*(9), 709-723.

Barnes, R. D., & Ivezaj, V. (2015). A systematic review of motivational interviewing for weight loss among adults in primary care. *Obesity Reviews*, *16*(4), 304-318.

Barrera, G. J., Sanchez, G., & Gonzalez, J. E. (2012). Trefoil factor 3 isolated from human breast milk downregulates cytokines (IL8 and IL6) and promotes human beta defensin (hBD2 and hBD4) expression in intestinal epithelial cells HT-29. *Bosnian Journal of Basic Medical Sciences*, *12*(4), 256.

Bartlett, M. S. (1954). A note on the multiplying factors for various $\chi 2$ approximations. *Journal of the Royal Statistical Society. Series B (Methodological)*, 296-298.

Basu, A., Wilkinson, M., Penugonda, K., Simmons, B., Betts, N. M., & Lyons, T. J. (2009). Freeze-dried strawberry powder improves lipid profile and lipid peroxidation in women with metabolic syndrome: baseline and post intervention effects. *Nutrition Journal*, *8*(1), 43.

Bates, B., Cox, L., Nicholson, S., Page, P., Prentice, A., Steer, T., & Swan, G. (2016). National Diet and Nutrition Survey results from years 5 and 6 (combined) of the Rolling Programme (2012/2013–2013/2014). *Public Health England:* London, UK.

Beard, J. L., & Connor, J. R. (2003). Iron status and neural functioning. *Annual Review of Nutrition, 23*(1), 41-58.

Beard, J., Borel, M., & Peterson, F. J. (1997). Changes in iron status during weight loss with very-low-energy diets. *American Journal of Clinical Nutrition*, *66*(1), 104-110.

Beck, K. L., Kruger, R., Conlon, C. A., Heath, A. L. M., Coad, J., Matthys, C., Jones, B. & Stonehouse, W. (2012). The relative validity and reproducibility of an iron food frequency questionnaire for identifying iron-related dietary patterns in young women. *Journal of the Academy of Nutrition and Dietetics*, *112*(8), 1177-1187.

Beck, K. L., Kruger, R., Conlon, C. A., Heath, A. L. M., Matthys, C., Coad, J., & Stonehouse, W. (2013). Suboptimal iron status and associated dietary patterns and practices in premenopausal women living in Auckland, New Zealand. *European Journal of Nutrition, 52*(2), 467-476.

Befort, C. A., Thomas, J. L., Daley, C. M., Rhode, P. C., & Ahluwalia, J. S. (2008). Perceptions and beliefs about body size, weight, and weight loss among obese African American women: a qualitative inquiry. *Health Education & Behavior, 35*(3), 410-426.

Bekri, S., Gual, P., Anty, R., Luciani, N., Dahman, M., Ramesh, B., Iannelli, A., Staccini-Myx, A., Casanova, D., Ben Amor, I., Saint-Paul, M.C., Huet, P.M., Sadoul, J.L., Gugenheim, J., Srai, S.K., Tran, A. & Le Marchand-Brustel, Y. (2006). Increased adipose tissue expression of hepcidin in severe obesity is independent from diabetes and NASH. *Gastroenterology*, *131*(3), 788-796.

Belza, B. L. (1995). Comparison of self-reported fatigue in rheumatoid arthritis and controls. *Journal of Rheumatology*, *22*(4), 639-643.

Berne, E. (1958). Transactional analysis: A new and effective method of group therapy. *American Journal of Psychotherapy*, *12*(4), 735-743.

Berne, E. (1961). *Transactional Analysis in Psychotherapy*. New York, Grove Press.

Bertinato, J., Aroche, C., Plouffe, L. J., Lee, M., Murtaza, Z., Kenney, L., Lavergne, C. & Aziz, A. (2014). Diet-induced obese rats have higher iron requirements and are more vulnerable to iron deficiency. *European Journal of Nutrition*, *53*(3), 885-895.

Bingham, S. A., Welch, A. A., McTaggart, A., Mulligan, A. A., Runswick, S. A., Luben, R., Suzy, O., Khaw, K.T., Wareham, N. & Day, N. E. (2001). Nutritional methods in the European prospective investigation of cancer in Norfolk. *Public Health Nutrition*, *4*(3), 847-858.

Braam, L. A. L. M., Ocke, M. C., Bueno-de-Mesquita, H. B., & Seidell, J. C. (1998). Determinants of obesity-related underreporting of energy intake. *American Journal of Epidemiology*, *147*(11), 1081-1086.

Braun, V., & Clarke, V. (2006). Using thematic analysis in psychology. *Qualitative Research in Psychology*, *3*(2), 77-101.

Bryant, M. J., Stevens, J., Truesdale, K. P., Mosley, T., & Chambless, L. (2008). Obesity and vital exhaustion: analysis of the atherosclerosis risk in the communities study. *Obesity*, *16*(7), 1545-1551.

Calder, P. C., Ahluwalia, N., Brouns, F., Buetler, T., Clement, K., Cunningham, K., Esposito, K., Jonsson, L. S., Kolb, H., Lansink, M., Marcos, A., Margioris,

A., Matusheski,N., Nordmann, H., O'Brien , J., Pugliese, G., Rizkalla, S., Schalkwijk, C., Tuomilehto, J., Julia Warnberg, J., Watzl, B., & Winklhofer-Roob, B. M. (2011). Dietary factors and low-grade inflammation in relation to overweight and obesity. *British Journal of Nutrition*, *106*(S3), 1-78.

Campbell, M. J., Machin, D., & Walters, S. J. (2010). *Medical statistics: a textbook for the health sciences.* New Jersey, John Wiley & Sons.

Canella, D. S., Levy, R. B., Martins, A. P. B., Claro, R. M., Moubarac, J. C., Baraldi, L. G., Moubarac, J.C., Baraldi, L.G., Cannon, G. & Monteiro, C. A. (2014). Ultra-processed food products and obesity in Brazilian households (2008–2009). *Plos One*, *9*(3), e92752.

Cani, P. D., Osto, M., Geurts, L., & Everard, A. (2012). Involvement of gut microbiota in the development of low-grade inflammation and type 2 diabetes associated with obesity. *Gut Microbes*, *3*(4), 279-288.

Cantor, J. B., Ashman, T., Gordon, W., Ginsberg, A., Engmann, C., Egan, M., Spielman, L., Dijkers, M. & Flanagan, S. (2008). Fatigue after traumatic brain injury and its impact on participation and quality of life. *Journal of Head Trauma Rehabilitation*, *23*(1), 41-51.

Carvalho, B. M., & Abdalla Saad, M. J. (2013). Influence of gut microbiota on subclinical inflammation and insulin resistance. *Mediators of Inflammation*, *2013*, 1-14.

Caselli, M., Cassol, F., Calò, G., Holton, J., Zuliani, G., & Gasbarrini, A. (2013). Actual concept of "probiotics": is it more functional to science or business. *World Journal of Gastroenterology*, *19*(10), 1527-1540.

Cattell, R. B. (1966). The scree test for the number of factors. *Multivariate Behavioral Research, 1*(2), 245-276.

Cepeda-Lopez, A. C., Melse-Boonstra, A., Zimmermann, M. B., & Herter-Aeberli, I. (2015). In overweight and obese women, dietary iron absorption is reduced and the enhancement of iron absorption by ascorbic acid is one-half that in normal-weight women. *American Journal of Clinical Nutrition*, *102*(6), 1389-1397.

Cepeda-Lopez, A. C., Osendarp, S. J., Melse-Boonstra, A., Aeberli, I., Gonzalez-Salazar, F., Feskens, E., Villalpando, S. & Zimmermann, M. B. (2011). Sharply higher rates of iron deficiency in obese Mexican women and children are predicted by obesity-related inflammation rather than by differences in dietary iron intake. *American Journal of Clinical Nutrition*, *93*(5), 975-983.

Cepeda-Lopez, A. C., Zimmermann, M. B., Wussler, S., Melse-Boonstra, A., Naef, N., Mueller, S. M., Toigo, M. & Herter-Aeberli, I. (2018). Greater blood volume and Hb mass in obese women quantified by the carbon monoxide-rebreathing method affects interpretation of iron biomarkers and iron requirements. *International Journal of Obesity*, *43*(5), 999–1008.

Cepeda-Lopez, A.C., Aeberli, I. & Zimmermann, M.B. (2010). Does obesity increase risk for iron deficiency? A review of the literature and the potential mechanisms. *International Journal for Vitamin and Nutrition Research, 80*(4), 263-270.

Chaiyasit, W., McClements, D. J., & Decker, E. A. (2005). The relationship between the physicochemical properties of antioxidants and their ability to

inhibit lipid oxidation in bulk oil and oil-in-water emulsions. *Journal of Agricultural and Food Chemistry*, *53*(12), 4982-4988.

Chambers, D., Phan, U., Chanadang, S., Maughan, C., Sanchez, K., Di Donfrancesco, B., Gomez, D., Higa, F., Li, H., Chambers, E. & Esen, E. (2016). Motivations for food consumption during specific eating occasions in Turkey. *Foods*, *5*(2), 39.

Cheng, H. L., Griffin, H. J., Bryant, C. E., Rooney, K. B., Steinbeck, K. S., & Helen, T. O. (2013). Impact of diet and weight loss on iron and zinc status in overweight and obese young women. *Asia Pacific Journal of Clinical Nutrition*, *22*(4), 574-582.

Cherayil, B. J. (2011). The role of iron in the immune response to bacterial infection. *Immunologic Research*, *50*(1), 1-9.

Cho, S., Dietrich, M., Brown, C. J., Clark, C. A., & Block, G. (2003). The effect of breakfast type on total daily energy intake and body mass index: results from the Third National Health and Nutrition Examination Survey (NHANES III). *Journal of the American College of Nutrition*, *22*(4), 296-302.

CIA. (2018). The World Factbook (Turkey). [ONLINE] Available at: https://www.cia.gov/library/publications/the-world-factbook/geos/tu.html. [Accessed 16 September 2018].

Clancy, S. M., Stroo, M., Schoenfisch, A., Dabrera, T., & Østbye, T. (2018). Barriers to engagement in a workplace weight management program: A qualitative study. *American Journal of Health Promotion, 32*(3), 763-770.

Coudray, C., Bellanger, J., Castiglia-Delavaud, C., Remesy, C., Vermorel, M., & Rayssignuier, Y. (1997). Effect of soluble or partly soluble dietary fibres supplementation on absorption and balance of calcium, magnesium, iron and zinc in healthy young men. *European Journal of Clinical Nutrition, 51*(6), 375.

Crane, T. E., Kubota, C., West, J. L., Kroggel, M. A., Wertheim, B. C., & Thomson, C. A. (2011). Increasing the vegetable intake dose is associated with a rise in plasma carotenoids without modifying oxidative stress or inflammation in overweight or obese postmenopausal women–3. *Journal of Nutrition*, *141*(10), 1827-1833.

Creswell, J. 2009. *Research design: Qualitative, quantitative, and mixed methods approaches.* California, SAGE Publications, Incorporated.

Crino, M., Sacks, G., Vandevijvere, S., Swinburn, B., & Neal, B. (2015). The influence on population weight gain and obesity of the macronutrient composition and energy density of the food supply. *Current Obesity Reports*, *4*(1), 1-10.

Damms-Machado, A., Weser, G., & Bischoff, S. C. (2012). Micronutrient deficiency in obese subjects undergoing low calorie diet. *Nutrition Journal*, *11*(1), 34.

Davies, M. B., & Hughes, N. (2014). *Doing a successful research project: Using qualitative or quantitative methods.* Basingstoke, Macmillan International Higher Education.

Department of Health. (1991). *Dietary Reference Values for Food Energy and Nutrients for the United Kingdom.* Richmond, HMSO.

Deeks, J. J., Dinnes, J., D'Amico, R., Sowden, A. J., Sakarovitch, C., Song, F., Petticrew, M. & Altman, D. G. (2003). Evaluating non-randomised intervention studies. *Health Technology Assessment (Winchester, England)*, *7*(27), iii-x.

Dien, J. (2010). Evaluating two-step PCA of ERP data with geomin, infomax, oblimin, promax, and varimax rotations. *Psychophysiology*, *47*(1), 170-183.

Ding, S., Chi, M. M., Scull, B. P., Rigby, R., Schwerbrock, N. M., Magness, S. & Lund, P. K. (2010). High-fat diet: bacteria interactions promote intestinal inflammation which precedes and correlates with obesity and insulin resistance in mouse. *Plos One*, *5*(8), e12191.

Downs, S. H., & Black, N. (1998). The feasibility of creating a checklist for the assessment of the methodological quality both of randomised and non-randomised studies of health care interventions. *Journal of Epidemiology & Community Health*, *52*(6), 377-384.

Eichelmann, F., Schwingshackl, L., Fedirko, V., & Aleksandrova, K. (2016). Effect of plant-based diets on obesity-related inflammatory profiles: a systematic review and meta-analysis of intervention trials. *Obesity Reviews*, *17*(11), 1067-1079.

El-Kader, S. M. A., Al-Jiffri, O. H., & Al-Shreef, F. M. (2015). Aerobic exercises alleviate symptoms of fatigue related to inflammatory cytokines in obese patients with type 2 diabetes. *African Health Sciences*, *15*(4), 1142-1148.

Englander, J., Bushnik, T., Oggins, J., & Katznelson, L. (2010). Fatigue after traumatic brain injury: Association with neuroendocrine, sleep, depression and other factors. *Brain Injury*, *24*(12), 1379-1388.

Fang, M. Z., Wang, Y., Ai, N., Hou, Z., Sun, Y., Lu, H., Welsh, W., & Yang, C. S. (2003). Tea polyphenol (−)-epigallocatechin-3-gallate inhibits DNA methyltransferase and reactivates methylation-silenced genes in cancer cell lines. *Cancer Research*, *63*(22), 7563-7570.

FAO. (2013). Food Balance Sheets. [ONLINE] Available at: http://www.fao.org/faostat/en/?#data/FBS. [Accessed 23 February 2019].

Faraj, M., Lavoie, M. È., Messier, L., Bastard, J. P., & Prud'homme, D. (2010). Reduction in serum apoB is associated with reduced inflammation and insulin resistance in post-menopausal women: A MONET study. *Atherosclerosis, 211*(2), 682-688.

Feng, R. N., Niu, Y. C., Sun, X. W., Li, Q., Zhao, C., Wang, C., Guo, F.C., Sun, C.H. & Li, Y. (2013). Histidine supplementation improves insulin resistance through suppressed inflammation in obese women with the metabolic syndrome: a randomised controlled trial. *Diabetologia, 56*(5), 985-994.

Fetters, M. D., Curry, L. A., & Creswell, J. W. (2013). Achieving integration in mixed methods designs—principles and practices. *Health Services Research, 48*(6pt2), 2134-2156.

Fuentes, E., Fuentes, F., Vilahur, G., Badimon, L. & Palomo, I. (2013). Mechanisms of chronic state of inflammation as mediators that link obese adipose tissue and metabolic syndrome. *Mediators of Inflammation*, 2013.

Fuller, N. R., Williams, K., Shrestha, R., Ahern, A. L., Holzapfel, C., Hauner, H., Jebb S.A. & Caterson, I. D. (2014). Changes in physical activity during a

weight loss intervention and follow-up: a randomized controlled trial. *Clinical Obesity, 4*(3), 127-135.

Fuqua, B. K., Vulpe, C. D., & Anderson, G. J. (2012). Intestinal iron absorption. *Journal of Trace Elements in Medicine and Biology, 26*(2-3), 115-119.

Ganz, T. (2005). Cellular iron: ferroportin is the only way out. *Cell Metabolism*, *1*(3), 155-157.

Ganz, T. (2015). Hepcidin and the global burden of iron deficiency. *Clinical Chemistry, 61*(4), 577-578.

Ganz, T., & Nemeth, E. (2012). Hepcidin and iron homeostasis. *Biochimica et Biophysica Acta (BBA)-Molecular Cell Research*, *1823*(9), 1434-1443.

Ganz, T., & Nemeth, E. (2015). Iron homeostasis in host defence and inflammation. *Nature Reviews Immunology*, *15*(8), 500-510.

Garip, G., & Yardley, L. (2011). A synthesis of qualitative research on overweight and obese people's views and experiences of weight management. *Clinical Obesity*, *1*(2-3), 110-126.

Geissler, C., & Singh, M. (2011). Iron, meat and health. *Nutrients*, *3*(3), 283-316.

Ghavipour, M., Saedisomeolia, A., Djalali, M., Sotoudeh, G., Eshraghyan, M. R., Moghadam, A. M., & Wood, L. G. (2013). Tomato juice consumption reduces systemic inflammation in overweight and obese females. *British Journal of Nutrition*, *109*(11), 2031-2035.

Gibson, R. S., Hotz, C., Temple, L., Yeudall, F., Mtitimuni, B., & Ferguson, E. (2000). Dietary strategies to combat deficiencies of iron, zinc, and vitamin A in developing countries: development, implementation, monitoring, and evaluation. *Food and Nutrition Bulletin*, *21*(2), 219-231.

Girelli, D., Nemeth, E., & Swinkels, D. W. (2016). Hepcidin in the diagnosis of iron disorders. *Blood*,*127*(23), 2809-2813..

Glesne, C. (2015). *Becoming qualitative researchers: An introduction*, 5th edition. Boston, Pearson.

Goodpaster, B. H., DeLany, J. P., Otto, A. D., Kuller, L., Vockley, J., South-Paul, J. E., Thomas S.B., Brown J., McTigue K., Hames K.C. & Lang, W. (2010). Effects of diet and physical activity interventions on weight loss and cardiometabolic risk factors in severely obese adults: a randomized trial. *Journal of the American Medical Association*, *304*(16), 1795-1802.

Grady, C., Anderson, R., & Chase, G. A. (1998). Fatigue in HIV-infected men receiving investigational interleukin-2. *Nursing Research*, *47*(4), 227-234.

Grant, K. E., Chandler, R. M., Castle, A. L., & Ivy, J. L. (1997). Chromium and exercise training: effect on obese women. *Medicine and Science in Sports and Exercise*, *29*(8), 992-998.

Greenhalgh, T., & Taylor, R. (1997). How to read a paper: papers that go beyond numbers (qualitative research). *British Medical Journal*, *315*(7110), 740-743.

Guest, D. D., Evans, E. M., & Rogers, L. Q. (2013). Diet components associated with perceived fatigue in breast cancer survivors. *European Journal of Cancer Care*, *22*(1), 51-59.

Guest, G., Bunce, A., & Johnson, L. (2006). How many interviews are enough? An experiment with data saturation and variability. *Field Methods*, *18*(1), 59-82.

Gulec, S., Anderson, G. J., & Collins, J. F. (2014). Mechanistic and regulatory aspects of intestinal iron absorption. *American Journal of Physiology-Gastrointestinal and Liver Physiology*, *307*(4), G397-G409.

Hacettepe University Institute of Population Studies. (2014). *2013 Turkey Demographic and Health Survey.* Hacettepe University Institute of Population Studies, T.R. Ministry of Development and TÜBİTAK, Ankara, Turkey.

Hassapidou, M., Fotiadou, E., Maglara, E., & Papadopoulou, S. K. (2006). Energy intake, diet composition, energy expenditure, and body fatness of adolescents in northern Greece. *Obesity*, *14*(5), 855-862.

Hawkes, C., Harris, J., & Gillespie, S. (2017). Changing diets: Urbanization and the nutrition transition. *International Food Policy Research Institute Book Chapters*, 34-41.Health and Social Care Information Centre. (2018). Statistics on obesity, physical activity and diet. [ONLINE] Available at: https://files.digital.nhs.uk/publication/0/0/obes-phys-acti-diet-eng-2018-rep.pdf. [Accessed 16 September 2018].

Herter-Aeberli, I., Thankachan, P., Bose, B., & Kurpad, A. V. (2015). Increased risk of iron deficiency and reduced iron absorption but no difference in zinc, vitamin A or B-vitamin status in obese women in India. *European Journal of Nutrition*, *55*(8), 2411-2421.

Horn, J. L. (1965). A rationale and test for the number of factors in factor analysis. *Psychometrika*, *30*(2), 179-185.

Hou, X., Shen, W., Huang, X., Ai, Z., & Zhang, L. (2016). Ascorbic acid enhanced activation of oxygen by ferrous iron: a case of aerobic degradation of rhodamine B. *Journal of Hazardous Materials*, *308*, 67-74.

Hurrell, R. F., Reddy, M., & Cook, J. D. (1999). Inhibition of non-haem iron absorption in man by polyphenolic-containing beverages. *British Journal of Nutrition*, *81*(4), 289-295.

Hurrell, R., & Egli, I. (2010). Iron bioavailability and dietary reference values–. *American Journal of Clinical Nutrition*, *91*(5), 1461S-1467S.

Hussein, L., Gouda, M., Fouad, M., Labib, E., Bassyouni, R., & Mohammad, M. (2014). Dietary intervention with yoghurt, Synbiotic yogurt or traditional fermented Sobya: bio-potency among male adolescents using five bio-markers of relevance to colonic metabolic activities. *Food and Nutrition Sciences*, *5*(12), 1131.

Impellizzeri, F. M., Agosti, F., De Col, A., & Sartorio, A. (2013). Psychometric properties of the Fatigue Severity Scale in obese patients. *Health and Quality of Life Outcomes*, *11*(1), 32.

International Physical Activity Questionnaire Research Committee. (2005). Guidelines for data processing and analysis of the international physical activity questionnaire (IPAQ)-short and long forms. [ONLINE] Available at: www.ipaq.ki.se [Accessed 16 September 2018].

Javadi, M., & Zarea, K. (2016). Understanding thematic analysis and its pitfall. *Journal of Client Care*, *1*(1), 33-39.

Jeffery, R. W., Wing, R. R., Sherwood, N. E., & Tate, D. F. (2003). Physical activity and weight loss: does prescribing higher physical activity goals improve outcome?. *American Journal of Clinical Nutrition*, *78*(4), 684-689.

Jiao, J., Xu, J. Y., Zhang, W., Han, S., & Qin, L. Q. (2015). Effect of dietary fiber on circulating C-reactive protein in overweight and obese adults: a meta-analysis of randomized controlled trials. *International Journal of Food Sciences and Nutrition*, *66*(1), 114-119.

Johns, D. J., Hartmann-Boyce, J., Jebb, S. A., Aveyard, P., & Behavioural Weight Management Review Group. (2014). Diet or exercise interventions vs combined behavioral weight management programs: a systematic review and meta-analysis of direct comparisons. *Journal of the Academy of Nutrition and Dietetics*, *114*(10), 1557-1568.

Jones, N., Furlanetto, D. L. C., Jackson, J. A., & Kinn, S. (2007). An investigation of obese adults' views of the outcomes of dietary treatment. *Journal of Human Nutrition and Dietetics*, *20*(5), 486-494.

Kaiser, H. F. (1970). A second generation little jiffy. *Psychometrika*, *35*(4), 401-415.

Kaiser, H. F. (1974). An index of factorial simplicity. *Psychometrika*, *39*(1), 31-36.

Kapil, R. (2017). Bioavailability & absorption of Iron and Anemia. *Indian Journal of Community Health*, *29*(4), 453-457.

Kazemzadeh, M., Safavi, S. M., Nematollahi, S., & Nourieh, Z. (2014). Effect of brown rice consumption on inflammatory marker and cardiovascular risk factors among overweight and obese non-menopausal female adults. *International Journal of Preventive Medicine*, *5*(4), 478.

Kelly, A. U., McSorley, S. T., Patel, P., & Talwar, D. (2017). Interpreting iron studies. *British Medical Journal, 357*, j2513.

Khan, A., Khan, W. M., Ayub, M., Humayun, M., & Haroon, M. (2016). Ferritin is a marker of inflammation rather than iron deficiency in overweight and obese people. *Journal of Obesity*, *2016*.

Knez, M., Nikolic, M., Zekovic, M., Stangoulis, J. C., Gurinovic, M., & Glibetic, M. (2017). The influence of food consumption and socio-economic factors on the relationship between zinc and iron intake and status in a healthy population. *Public Health Nutrition*, *20*(14), 2486-2498.

Kowal, J., & Fortier, M. S. (2007). Physical activity behavior change in middle-aged and older women: the role of barriers and of environmental characteristics. *Journal of Behavioral Medicine*, *30*(3), 233-242.

Krayenbuehl, P. A., Battegay, E., Breymann, C., Furrer, J., & Schulthess, G. (2011). Intravenous iron for the treatment of fatigue in non-anemic,

premenopausal women with low serum ferritin concentration. *Blood*, *118*(12), 3222-3227

Kretsch, M. J., Fong, A. K. H., Green, M. W., & Johnson, H. L. (1998). Cognitive function, iron status, and hemoglobin concentration in obese dieting women. *European Journal of Clinical Nutrition*, *52*(7), 512.

Labonté, M. È., Couture, P., Richard, C., Desroches, S., & Lamarche, B. (2013). Impact of dairy products on biomarkers of inflammation: a systematic review of randomized controlled nutritional intervention studies in overweight and obese adults. *American Journal of Clinical Nutrition*, *97*(4), 706-717.

Lane, D. J.R., & Richardson, D. R. (2014). The active role of vitamin C in mammalian iron metabolism: much more than just enhanced iron absorption!. *Free Radical Biology and Medicine*, *75*, 69-83.

Lane, D. J.R., Chikhani, S., Richardson, V., & Richardson, D. R. (2013). Transferrin iron uptake is stimulated by ascorbate via an intracellular reductive mechanism. *Biochimica et Biophysica Acta (BBA)-Molecular Cell Research*, *1833*(6), 1527-1541.

Lasselin, J., Layé, S., Dexpert, S., Aubert, A., Gonzalez, C., Gin, H., & Capuron, L. (2012). Fatigue symptoms relate to systemic inflammation in patients with type 2 diabetes. *Brain, Behavior, and Immunity*, *26*(8), 1211-1219.

Lasselin, J., & Capuron, L. (2014). Chronic low-grade inflammation in metabolic disorders: relevance for behavioral symptoms. *Neuroimmunomodulation*, *21*(2-3), 95-101.

Lemstra, M., & Rogers, M. R. (2015). The importance of community consultation and social support in adhering to an obesity reduction program: results from the Healthy Weights Initiative. *Patient Preference and Adherence*, *9*, 1473.

Lim, K. H., Riddell, L. J., Nowson, C. A., Booth, A. O., & Szymlek-Gay, E. A. (2013). Iron and zinc nutrition in the economically-developed world: a review. *Nutrients*, *5*(8), 3184-3211.

Lim, W., Hong, S., Nelesen, R., & Dimsdale, J. E. (2005). The association of obesity, cytokine levels, and depressive symptoms with diverse measures of fatigue in healthy subjects. *Archives of Internal Medicine*, *165*(8), 910-915.

Lopez, A., Cacoub, P., Macdougall, I. C., & Peyrin-Biroulet, L. (2016). Iron deficiency anaemia. *Lancet*, *387*(10021), 907-916.

Louzada, M. L., Baraldi, L. G., Steele, E. M., Martins, A. P. B., Canella, D. S., Moubarac, J. C., Levy, R.B., Cannon, G., Afshin, A., Imamura, F., Mozaffarian, D. & Monteiro, C.A. (2015). Consumption of ultra-processed foods and obesity in Brazilian adolescents and adults. *Preventive Medicine*, *81*, 9-15.

Lüscher, T. F. (2015). Ageing, inflammation, and oxidative stress: final common pathways of cardiovascular disease. *European Heart Journal,* 36(48), 3381-33383.

Ma, Q., Kim, E. Y., & Han, O. (2010). Bioactive dietary polyphenols decrease heme iron absorption by decreasing basolateral iron release in human intestinal Caco-2 cells. *Journal of Nutrition*, *140*(6), 1117-1121.

Maffiuletti, N. A., Jubeau, M., Munzinger, U., Bizzini, M., Agosti, F., De Col, A., Lafortuna, C.L. & Sartorio, A. (2007). Differences in quadriceps muscle strength and fatigue between lean and obese subjects. *European Journal of Applied Physiology*, *101*(1), 51-59.

Maguire, M., & Delahunt, B. (2017). Doing a thematic analysis: A practical, step-by-step guide for learning and teaching scholars. *All Ireland Journal of Teaching and Learning in Higher Education*, *9*(3), 3351-3364.

Mascitelli, L., & Goldstein, M. R. (2014). Inhibition of iron absorption by polyphenols as an anticancer mechanism. *Polyphenols in Human Health and Disease* (pp. 1283-1286).

McClung, J.P. & Karl, J.P. (2009). Iron deficiency and obesity: the contribution of inflammation and diminished iron absorption. *Nutrition Reviews*, 67(2), 100-104.

McCusker, K., & Gunaydin, S. (2015). Research using qualitative, quantitative or mixed methods and choice based on the research. *Perfusion*, *30*(7), 537-542.

Mehta, R. K. (2015). Impacts of obesity and stress on neuromuscular fatigue development and associated heart rate variability. *International Journal of Obesity*, *39*(2), 208.

Mendonça, R. D. D., Pimenta, A. M., Gea, A., de la Fuente-Arrillaga, C., Martinez-Gonzalez, M. A., Lopes, A. C. S., & Bes-Rastrollo, M. (2016). Ultraprocessed food consumption and risk of overweight and obesity: the University of Navarra Follow-Up (SUN) cohort study. *American Journal of Clinical Nutrition*, *104*(5), 1433-1440.

Mennen, L., Hirvonen, T., Arnault, N., Bertrais, S., Galan, P., & Hercberg, S. (2007). Consumption of black, green and herbal tea and iron status in French adults. *European Journal of Clinical Nutrition*, *61*(10), 1174.

Miller, W. R., & Rollnick, S. (2012). *Motivational interviewing: Helping people change*. New York, Guilford press.

Ministry of Health of Turkey. (2010). Obesity prevention and control program of Turkey (2010-2014). [ONLINE] Available at: http://web.archive.org/web/20160802174123/http://beslenme.gov.tr/content/files/home/obesity_prevention_and_control_program_of_turkey_2010_2014.pdf (Accessed 23 February 2019)

Ministry of Health of Turkey. (2017). Diyetisyenler icin hasta izlem rehberi, agirlik yonetimi el kitabi. Ankara, Turkey

Moll, R., & Davis, B. (2017). Iron, vitamin B12 and folate. *Medicine*, *45*(4), 198-203.

Morgan, D.L. (1996). Focus groups. *Annual Review of Sociology*, *22*(1),129-152.

Moschonis, G., Chrousos, G. P., Lionis, C., Mougios, V., & Manios, Y. (2012). Association of total body and visceral fat mass with iron deficiency in preadolescents: The Healthy Growth Study. *British Journal of Nutrition*, *108*(04), 710-719.

Moubarac, J. C., Batal, M., Louzada, M. L., Steele, E. M., & Monteiro, C. A. (2017). Consumption of ultra-processed foods predicts diet quality in Canada. *Appetite*, *108*, 512-520.

Mujica-Coopman, M. F., Brito, A., de Romana, D. L., Pizarro, F., & Olivares, M. (2015). Body mass index, iron absorption and iron status in childbearing age women. *Journal of Trace Elements in Medicine and Biology, 30*, 215-219.

Nair, K. M., & Augustine, L. F. (2018). Food synergies for improving bioavailability of micronutrients from plant foods. *Food Chemistry*, *238*, 180-185.

Nemeth, E., & Ganz, T. (2014). Anemia of inflammation. *Hematology/Oncology Clinics*, *28*(4), 671-681.

Neuberger, G. B. (2003). Measures of fatigue: the fatigue questionnaire, fatigue severity scale, multidimensional assessment of fatigue scale, and short form-36 vitality (energy/fatigue) subscale of the short form health survey. *Arthritis Care & Research: Official Journal of the American College of Rheumatology*, *49*(S5), 175-183.

Nunnally, J. C., & Bernstein, I. H. (1978). *Psychometric Theory*. New York, McGraw-Hill.

OECD. 2017. Obesity update. [ONLINE] Available at: http://www.oecd.org/health/health-systems/Obesity-Update-2017.pdf. (Accessed 16 September 2018).

Pallant, J., (2016). *SPSS survival manual: a step by step guide to data analysis using IBM SPSS.* 6th ed., Maidenhead, Open University Press.

Papanikolaou, G., & Pantopoulos, K. (2005). Iron metabolism and toxicity. *Toxicology and Applied Pharmacology*, *202*(2), 199-211.

Papanikolaou, G., & Pantopoulos, K. (2017). Systemic iron homeostasis and erythropoiesis. J*ournal of the International Union of Biochemistry and Molecular Biology Life*, *69*(6), 399-413.

Pasricha, S. R., Drakesmith, H., Black, J., Hipgrave, D., & Biggs, B. A. (2013). Control of iron deficiency anemia in low-and middle-income countries. *Blood*, *121*(14), 2607-2617.

Patterson, A. J., Brown, W. J., & Roberts, D. C. (2001). Dietary and supplement treatment of iron deficiency results in improvements in general health and fatigue in Australian women of childbearing age. *Journal of the American College of Nutrition*, *20*(4), 337-342.

Petry, N., Olofin, I., Hurrell, R. F., Boy, E., Wirth, J. P., Moursi, M., Angel, D.M. & Rohner, F. (2016). The proportion of anemia associated with iron deficiency in low, medium, and high human development index countries: a systematic analysis of national surveys. *Nutrients*, *8*(11), 693.

Pfeiffer, C. M., & Looker, A. C. (2017). Laboratory methodologies for indicators of iron status: strengths, limitations, and analytical challenges. *American Journal of Clinical Nutrition*, *106*(6), 1606S-1614S.

Pinstrup-Andersen, P. (2007). Agricultural research and policy for better health and nutrition in developing countries: a food systems approach. *Agricultural Economics*, *37*, 187-198.

Piper, B. F., Dibble, S. L., Dodd, M. J., Weiss, M. C., Slaughter, R. E., & Paul, S. M. (1998). The revised Piper Fatigue Scale: psychometric evaluation in women with breast cancer. *Oncology Nursing Forum*, *25*, 677-684.

Pizarro, F., Olivares, M., Valenzuela, C., Brito, A., Weinborn, V., Flores, S., & Arredondo, M. (2016). The effect of proteins from animal source foods on heme iron bioavailability in humans. *Food Chemistry*, *196,* 733-738.

Prystai, E. A., Kies, C. V., & Driskell, J. A. (1999). Calcium, copper, iron, magnesium and zinc utilization of humans as affected by consumption of black, decaffeinated black and green teas. *Nutrition Research*, *19*(2), 167-177.

QSR International. 2018. *QSR NVivo Version 12* [ONLINE]. Available: http://www.qsrinternational.com (Accessed 7 August 2019).

Randell, E., McNamara, R., Shaw, C., Espinasse, A., & Simpson, S. A. (2015). Challenges of a community based pragmatic, randomised controlled trial of weight loss maintenance. *Biomed Central Research Notes*, *8*(1), 802.

Reddy, M. B., Hurrell, R. F., & Cook, J. D. (2006). Meat consumption in a varied diet marginally influences nonheme iron absorption in normal individuals. *Journal of Nutrition*, *136*(3), 576-581.

Ren, G. Y., Chen, C. Y., Chen, G. C., Chen, W. G., Pan, A., Pan, C. W., Zhang, Y.H., Qin, L.Q. & Chen, L. H. (2016). Effect of flaxseed intervention on

inflammatory marker c-reactive protein: a systematic review and meta-analysis of randomized controlled trials. *Nutrients*, *8*(3), 136.

Roberts, C., Steer, T., Maplethorpe, N., Cox, L., Meadows, S., Nicholson, S., Page, P. & Swan, G. (2018). National Diet and Nutrition Survey: results from years 7 and 8 (combined) of the Rolling Programme (2014/2015–2015/2016).

Rodriguez-Rodriguez, E., Lopez-Sobaler, A. M., Andres, P., Aparicio, A., Navia, B., & Ortega, R. M. (2007). Modification of iron status in young overweight/mildly obese women by two dietary interventions designed to achieve weight loss. *Annals of Nutrition and Metabolism*, *51*(4), 367-373.

Rosado, J. L., Garcia, O. P., Ronquillo, D., Hervert-Hernández, D., Caamaño, M. D. C., Martínez, G., Gutiérrez, J. & García, S. (2011). Intake of milk with added micronutrients increases the effectiveness of an energy-restricted diet to reduce body weight: a randomized controlled clinical trial in Mexican women. *Journal of the American Dietetic Association*, *111*(10), 1507-1516.

Ryan, A. S., Ge, S., Blumenthal, J. B., Serra, M. C., Prior, S. J., & Goldberg, A. P. (2014). Aerobic exercise and weight loss reduce vascular markers of inflammation and improve insulin sensitivity in obese women. *Journal of the American Geriatrics Society*, *62*(4), 607-614.

Saito, H., Kimura, Y., Tashima, S., Takao, N., Nakagawa, A., Baba, T., & Sato, S. (2009). Psychological factors that promote behavior modification by obese patients. *BioPsychoSocial medicine*, *3*(1), 9.

Samman, S., Sandström, B., Toft, M. B., Bukhave, K., Jensen, M., Sørensen, S. S., & Hansen, M. (2001). Green tea or rosemary extract added to foods reduces

nonheme-iron absorption. *American Journal of Clinical Nutrition*, *73*(3), 607-612.

Sandelowski, M. (1995). Sample size in qualitative research. *Research in Nursing & Health*, *18*(2), 179-183.

Sawada, T., Konomi, A., & Yokoi, K. (2014). Iron deficiency without anemia is associated with anger and fatigue in young Japanese women. *Biological Trace Element Research*, *159*(1-3), 22-31.

Saydam, B. K., Genc, R. E., Sarac, F., & Turfan, E. C. (2017). Prevalence of anemia and related factors among women in Turkey. *Pakistan Journal of Medical Sciences*, *33*(2), 433.

Scheers, N., Rossander-Hulthen, L., Torsdottir, I., & Sandberg, A. S. (2016). Increased iron bioavailability from lactic-fermented vegetables is likely an effect of promoting the formation of ferric iron (Fe 3+). *European Journal of Nutrition*, *55*(1), 373-382.

Schmidt, P. J. (2015). Regulation of iron metabolism by hepcidin under conditions of inflammation. *Journal of Biological Chemistry*, *290*(31), 18975-18983.

Scientific Advisory Committee on Nutrition. (2010). SCAN Iron and Health Report. [ONLINE] Available at: https://www.gov.uk/government/publications/sacn-iron-and-health-report (Accessed 7 August 2019).

Selvin, E., Paynter, N. P., & Erlinger, T. P. (2007). The effect of weight loss on C-reactive protein: a systematic review. *Archives of Internal Medicine*, *167*(1), 31-39.

Sharp, P. A. (2010). Intestinal iron absorption: regulation by dietary & systemic factors. *International Journal for Vitamin and Nutrition Research*, *80* (4-5), 231-242.

Shewfelt, R. L. (2017). How Does Food Processing Change the Nutritional Value of Foods?. *In Defense of Processed Food* (pp. 107-123). Copernicus, Cham.

Shoelson, S.E. & Goldfine, A.B. (2009). Getting away from glucose: fanning the flames of obesity-induced inflammation. *Nature Medicine, 15*(4), 373-374.

Shrivastava, A. K., Singh, H. V., Raizada, A., & Singh, S. K. (2015). C-reactive protein, inflammation and coronary heart disease. *Egyptian Heart Journal*, *67*(2), 89-97.

Silva, B., & Faustino, P. (2015). An overview of molecular basis of iron metabolism regulation and the associated pathologies. *Biochimica et Biophysica Acta (BBA)-Molecular Basis of Disease*, *1852*(7), 1347-1359.

Simpson, K. A., & Singh, M. A. F. (2008). Effects of exercise on adiponectin: a systematic review. *Obesity*, *16*(2), 241-256.

Singh, P., & Prasad, S. (2018). Determination of ascorbic acid and its influence on the bioavailability of iron, zinc and calcium in Fijian food samples. *Microchemical Journal*, *139*, 119-124.

Slimming World. (2018). About Slimming World. [ONLINE] Available at: https://www.slimmingworld.co.uk/health/how-sw-works/about-slimming-world.aspx. (Accessed 16 September 2018).

Slimming World. (2019). Facilitating behaviour change to tackle obesity. [ONLINE] Available at: https://www.slimmingworld.co.uk/health/downloads/sw-behaviour-change.pdf (Accessed 15 March 2019).

Smith, J., & Firth, J. (2011). Qualitative data analysis: the framework approach. *Nurse Researcher*, *18*(2), 52-62.

Statista. (2019). Annual per capita tea consumption worldwide as of 2016, by leading countries (in pounds). [ONLINE] Available at: https://www.statista.com/statistics/507950/global-per-capita-tea-consumption-by-country (Accessed 25 March 2019).

Stubbs, J., Brogelli, D., Pallister, C., Avery, A., McConnon, A., & Lavin, J. (2012). Behavioural and motivational factors associated with weight loss and maintenance in a commercial weight management programme. *The Open Obesity Journal*, *4*(1), 35-43.

Suchdev, P. S., Williams, A. M., Mei, Z., Flores-Ayala, R., Pasricha, S. R., Rogers, L. M., & Namaste, S. M. (2017). Assessment of iron status in settings of inflammation: challenges and potential approaches. *American Journal of Clinical Nutrition*, *106*(suppl_6), 1626S-1633S.

Swift, J. A., & Tischler, V. (2010). Qualitative research in nutrition and dietetics: getting started. *Journal of Human Nutrition and Dietetics*, *23*(6), 559-566.

Tabachnik, B. G., & Fidell, L. S. (2013). *Using multivariate statistics (6e éd.)*. Boston, Pearson.

Temme, E. H. M., & Van Hoydonck, P. G. A. (2002). Tea consumption and iron status. *European Journal of Clinical Nutrition*, *56*(5), 379.

Thankachan, P., Walczyk, T., Muthayya, S., Kurpad, A. V., & Hurrell, R. F. (2008). Iron absorption in young Indian women: the interaction of iron status with the influence of tea and ascorbic acid. *American Journal of Clinical Nutrition*, *87*(4), 881-886.

Thomas, S. L., Hyde, J., Karunaratne, A., Kausman, R., & Komesaroff, P. A. (2008). " They all work... when you stick to them": A qualitative investigation of dieting, weight loss, and physical exercise, in obese individuals. *Nutrition Journal*, *7*(1), 34.

Thongprasert, C., Hutchinson, C., Satheannoppakao, W., & Tipayamongkholgul, M. (2017). Dietary iron intake and availability are related to maternal education level in overweight/obese adolescents. *European Journal of Nutrition*, *57*(6), 2249-2260.

Thorp, A. A., Kingwell, B. A., Owen, N., & Dunstan, D. W. (2014). Breaking up workplace sitting time with intermittent standing bouts improves fatigue and musculoskeletal discomfort in overweight/obese office workers. *Occupational and Environmental Medicine*, *71*(11), 765-771.

Timmerman, G. M. (1999). Using self-care strategies to make lifestyle changes. *Journal of Holistic Nursing*, *17*(2), 169-183.

Turan, Y., Duruöz, M. T., Bal, S., Guvenc, A., Cerrahoglu, L., & Gurgan, A. (2007). Assessment of fatigue in patients with ankylosing spondylitis. *Rheumatology International*, *27*(9), 847-852.

Tussing-Humphreys, L. M., Nemeth, E., Fantuzzi, G., Freels, S., Guzman, G., Holterman, A. X. L., & Braunschweig, C. (2010a). Elevated systemic hepcidin and iron depletion in obese premenopausal females. *Obesity*, *18*(7), 1449-1456.

Tussing-Humphreys, L. M., Nemeth, E., Fantuzzi, G., Freels, S., Holterman, A. X. L., Galvani, C., Ayloo, S., Vitello, J. & Braunschweig, C. (2010b). Decreased serum hepcidin and improved functional iron status 6 months after restrictive bariatric surgery. *Obesity*, *18*(10), 2010-2016.

Tussing-Humphreys, L., Pustacioglu, C., Nemeth, E. & Braunschweig, C. (2012). Rethinking iron regulation and assessment in iron deficiency, anemia of chronic disease, and obesity: introducing hepcidin. *Journal of the Academy of Nutrition and Dietetics, 112*(3), 391-400.

Vinokurov, E., & Libman, A. (2012). *Eurasian integration: Challenges of transcontinental regionalism*. Basingstoke, Palgrave Macmillan.

Volpe, S. L., Huang, H. W., Larpadisorn, K., & Lesser, I. I. (2001). Effect of chromium supplementation and exercise on body composition, resting metabolic rate and selected biochemical parameters in moderately obese women following an exercise program. *Journal of the American College of Nutrition*, *20*(4), 293-306.

Waldvogel-Abramowski, S., Waeber, G., Gassner, C., Buser, A., Frey, B.M., Favrat, B. & Tissot, J.D. (2014). Physiology of iron metabolism. *Transfusion Medicine and Hemotherapy*, 41(3), 213-221.

Wallace, D. F. (2016). The regulation of iron absorption and homeostasis. *Clinical Biochemist Reviews*, *37*(2), 51-62.

Wang, J., Sereika, S. M., Chasens, E. R., Ewing, L. J., Matthews, J. T., & Burke, L. E. (2012). Effect of adherence to self-monitoring of diet and physical activity on weight loss in a technology-supported behavioral intervention. *Patient Preference and Adherence*, *6*, 221.

Wang, Z. & Nakayama, T. (2010). Inflammation, a link between obesity and cardiovascular disease. *Mediators of Inflammation*. 2010, 1-17.

Watanabe, M., Suliman, M. E., Qureshi, A. R., Garcia-Lopez, E., Bárány, P., Heimbürger, O., Stenvinkel, P. & Lindholm, B. (2008). Consequences of low plasma histidine in chronic kidney disease patients: associations with inflammation, oxidative stress, and mortality. *American Journal of Clinical Nutrition*, *87*(6), 1860-1866.

Watkins, M. W. (2000). Monte Carlo PCA for parallel analysis [computer software]. [ONLINE] Available at: http://espse.ed.psu.edu/spsy/watkins/watkins3.ssi (Accessed 25 April 2018)

Weinborn, V., Pizarro, F., Olivares, M., Brito, A., Arredondo, M., Flores, S., & Valenzuela, C. (2015). The Effect of Plant Proteins Derived from Cereals and Legumes on Heme Iron Absorption. *Nutrients*, *7*(11), 8977-8986.

Weinborn, V., Valenzuela, C., Olivares, M., Arredondo, M., Weill, R., & Pizarro, F. (2017). Prebiotics increase heme iron bioavailability and do not affect non-heme iron bioavailability in humans. *Food & Function*, *8*(5), 1994-1999.

Weiss, G. (2009). Iron metabolism in the anemia of chronic disease. *Biochimica et Biophysica Acta (BBA)-General Subjects*, *1790*(7), 682-693.

Wenzel, B., Stults, H., & Mayer, J. (1962). Hypoferraemia in obese adolescents. *Lancet*, *2*, 327-328.

WHO. (2001). Iron deficiency anemia. Assessment, prevention, and control. *A guide for programme managers*, 47-62. [ONLINE] Available at: https://www.who.int/nutrition/publications/micronutrients/anaemia_iron_deficiency/WHO_NHD_01.3/en/ (Accessed 16 September 2018).

WHO. (2012). Resolution WHA65.6. Maternal, infant and young child nutrition. In: Sixty-fifth World Health Assembly, Geneva, 21–26 May. Resolutions and Decisions, Annexes. World Health Organization: Geneva.

WHO. (2013). Global action plan for the prevention and control of noncommunicable diseases 2013-2020. [ONLINE] Available at: http://africahealthforum.afro.who.int/IMG/pdf/global_action_plan_for_the_prevention_and_control_of_ncds_2013-2020.pdf. (Accessed 16 September 2018).

WHO. (2014). Comprehensive implementation plan on maternal, infant and young child nutrition. [ONLINE] Available at: https://apps.who.int/iris/bitstream/handle/10665/113048/WHO_NMH_NHD_14.1_eng.pdf;jsessionid=0B74BEF44B3F2189EC92EA8420B26862?sequence=1 (Accessed 16 September 2018).

WHO. (2014). Global nutrition targets 2025: anaemia policy brief. [ONLINE] Available at: https://www.who.int/nutrition/publications/globaltargets2025_policybrief_anaemia/en/ (Accessed 16 September 2018).

WHO. (2015). The global prevalence of anaemia in 2011. [ONLINE] Available at: https://www.who.int/nutrition/publications/micronutrients/global_prevalence_anaemia_2011/en/ (Accessed 16 September 2018).

WHO. (2016). Micronutrient deficiencies. [ONLINE] Available at: http://www.who.int/nutrition/topics/ida/en/ (Accessed: 15 Feb 2016).

WHO. (2018). National household health survey in turkey: prevalence of noncommunicable disease risk factors, 2017. [ONLINE] Available at: http://www.euro.who.int/en/countries/turkey/publications/national-household-health-survey-prevalence-of-noncommunicable-disease-risk-factors-in-turkey-2017-2018 (Accessed 16 September 2018).

WHO. (2018). Obesity and overweight. [ONLINE] Available at: http://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight. (Accessed 16 September 2018).

Wuensch, K. L. (2012). Principal component analysis-SPSS. [ONLINE] Available at: core.ecu.edu/psyc/wuenschk/MV/FA/PCA-SPSS.docx (Accessed 25 April 2018).

Wyatt, H. R., Grunwald, G. K., Mosca, C. L., Klem, M. L., Wing, R. R., & Hill, J. O. (2002). Long-term weight loss and breakfast in subjects in the National Weight Control Registry. *Obesity Research*, *10*(2), 78-82.

Xiao, S., Fei, N., Pang, X., Shen, J., Wang, L., Zhang, B., & Sun, L. (2014). A gut microbiota-targeted dietary intervention for amelioration of chronic inflammation underlying metabolic syndrome. *Journal of The Federation of European Microbiological Societies, 87*(2), 357-367.

Yanoff, L.B., Menzie, C.M., Denkinger, B., Sebring, N.G., McHugh, T., Remaley, A.T. & Yanovski, J.A. (2007). Inflammation and iron deficiency in the hypoferremia of obesity. *International Journal of Obesity*, *31*(9), 1412-1419.

Yildirim, Y., & Ergin, G. (2013). A validity and reliability study of the Turkish Multidimensional Assessment of Fatigue (MAF) scale in chronic musculoskeletal physical therapy patients. *Journal of Back and Musculoskeletal Rehabilitation*, *26*(3), 307-316.

Zijp, I. M., Korver, O., & Tijburg, L. B. (2000). Effect of tea and other dietary factors on iron absorption. *Critical Reviews in Food Science and Nutrition*, *40*(5), 371-398.

Zimmermann, M.B., Zeder, C., Muthayya, S., Winichagoon, P., Chaouki, N., Aeberli, I. & Hurrell, R.F. (2008). Adiposity in women and children from transition countries predicts decreased iron absorption, iron deficiency and a reduced response to iron fortification. *International Journal of Obesity, 32*(7), 1098-1104.

APPENDIX A: Abstract for oral presentation at the International Conference on Obesity and Diabetes, July 10-12, 2017, Dubai, UAE

Title: Does Engagement in a Weight Management Programme Improve Level of Fatigue?

Scientific Track: Nutritional Sciences

Hüsna Kaya Kaçar
Co-authors: Fiona McCullough
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Introduction: Obesity, where body mass index (BMI) ≥30 kg/m^2, is a major public health issue affecting approximately 25% of women in England[1]. People with obesity tend to have poorer sleep quality, a higher level of fatigue, and lower level of physical activity than healthy weight individuals[2]. Engagement in a weight management programme might be a way to address these obesity-related associations. This study aims to investigate effects of a weight management programme on level of fatigue in women of child-bearing age.

Methods: An online survey completed by non-pregnant female new members of Slimming World. The questionnaire asks for before joining (T0) and current data (T1), including weight, height, and level of fatigue experienced, using validated questions (Multidimensional Assessment of Fatigue, MAPI Research Trust[3]). 12 weeks (T2) later, the same participants were invited via e-mail to complete the second survey using the same questions. Global fatigue index (GFI) was calculated for each time point. Ethical approval was received.

Results: Completed data for T0 and T1 for 543 participants and for T2, n=75. Early analysis indicated that there were significant differences ($p<0.001$) between T0-T1, and T1-T2 in terms of BMI and level of fatigue.

Discussion/Conclusion: Engagement in a weight management programme is successful in reducing level of fatigue in women with obesity. Further analyses will be undertaken to establish whether there might be a correlation between weight loss and level of fatigue and whether changes in dietary habits may contribute to both outcome measures.

References:

1. Public Health England. (2016). Adult obesity international comparisons data factsheet
2. Jarosz, P. A., Davis, J. E., Yarandi, H. N., Farkas, R., Feingold, E., Shippings, S. H., Arlanda, L.S. & Williams, D. (2014). Obesity in urban women: associations with sleep and sleepiness, fatigue and activity. *Women's Health Issues*, *24*(4), e447-e454.
3. Belza, B. L., Henke, C. J., Yelin, E. H., Epstein, W. V., & Gilliss, C. L. (1993). Correlates of fatigue in older adults with rheumatoid arthritis. Nursing research, 42(2), 93-99.

APPENDIX B: Abstract for oral presentation at Women and Maternal Nutrition and Care, August 31-September 01, 2018, Toronto, Canada

Husna Kaya Kaçar et al., J Nutr Food Sci 2018, Volume 8
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Annual Conference on
Women and Maternal Nutrition and Care
Aug 31-Sep 01, 2018 | Toronto, Canada

Associations between body weight, fatigue and levels of physical activity in non-pregnant women accessing a weight management programme

Husna Kaya Kaçar and Fiona McCullough
University of Nottingham, UK

Individuals with obesity have higher levels of fatigue than people of healthy weight. Weight management programmes encourage participants to improve both their diet and physical activity levels. This study investigates the effects of a weight management programme (SW) on weight loss, physical activity and level of fatigue with the aim to understand the associations between weight change and levels of fatigue and physical activity. An online survey, hosted by Slimming World, completed by new female adult members. The questionnaire included before joining (T0) and current data (T1): weight, height, level of fatigue (using the validated Multidimensional Assessment of Fatigue) and level of physical activity (IPAQ-short form). 12 weeks (T2) later, the same participants were invited to complete a second survey asking the same questions. The association between weight status, fatigue level and physical activity level were investigated by Multiple Regression models.74 eligible women (BMI≥30kg/m2) completed the questionnaire at all time-points. The analysis indicated significant differences ($p<0.001$) over the three-time points in terms of BMI, physical activity and fatigue levels. A significant positive correlation between weight loss and a decrease in fatigue level was found ($r=.27$, $p<.05$). Also, physical activity level at T2 was significantly and positively correlated with weight loss ($r=.24$, $p<.05$). A weight management programme which encourages healthy lifestyle changes may help to reduce levels of fatigue in females with obesity. This may potentially enable the women to maintain the lifestyle changes.

Biography

Kaya Kacar is currently doing Ph.D. in Nutritional Sciences at the University of Nottingham, UK. She received her bachelor's degree from the University of Erciyes, Turkey. She then worked in a public hospital in Turkey, served as a dietitian. She completed her Master in Advanced Dietetic Practice from the University of Nottingham. She is a fellow in Turkish Dietetic Association. She is awarded by Turkish Ministry of Education Ph.D. scholarship.

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Notes:

APPENDIX C: Abstract for poster presentation at the 17th International Congress of Dietetics, September 7-10, 2016, Granada, Spain

Iron deficiency and Obesity

Introduction

Hidden hunger is a global public health concern with iron and other nutritional deficiencies masked by signs of over nutrition.

The inverse relationship between iron status and obesity was first reported by Wenzel et al. (1962) and over recent years there has been renewed interest. Research by Kaner (2014) found the prevalence of iron deficiency and iron deficiency anaemia in the obese, overweight and normal weight in women in Turkey were 45.6%, 27.4%, 41.9%, 24.0%, 23.5%, 12.4%, respectively.

Reasons why obese individuals are more likely to be iron deficient include inadequate dietary iron intake and increased iron requirements because of increased blood volume (Failla et al., 1988). More recent studies have suggested impaired duodenal iron absorption, obesity-associated low-grade inflammation influencing iron status and alterations to the iron regulatory protein hepcidin.

There is no guidance on the best means of treatment to improve iron status in the obese person and whether recommended dietary iron intake should be increased to compensate for alterations to the iron regulatory processes.

The aim of the study is to evaluate interventions which treat iron deficiency anaemia in obese individuals, particularly in women of reproductive age, with a view to informing public health recommendations in Turkey.

Methods

A systematic review will be undertaken to assess the effectiveness of interventions designed to address iron deficiency among women of reproductive age who are either overweight or obese. The findings will be used to influence the development and feasibility of an intervention to be delivered to the same population group in Turkey with the primary outcome measure being level of iron deficiency.

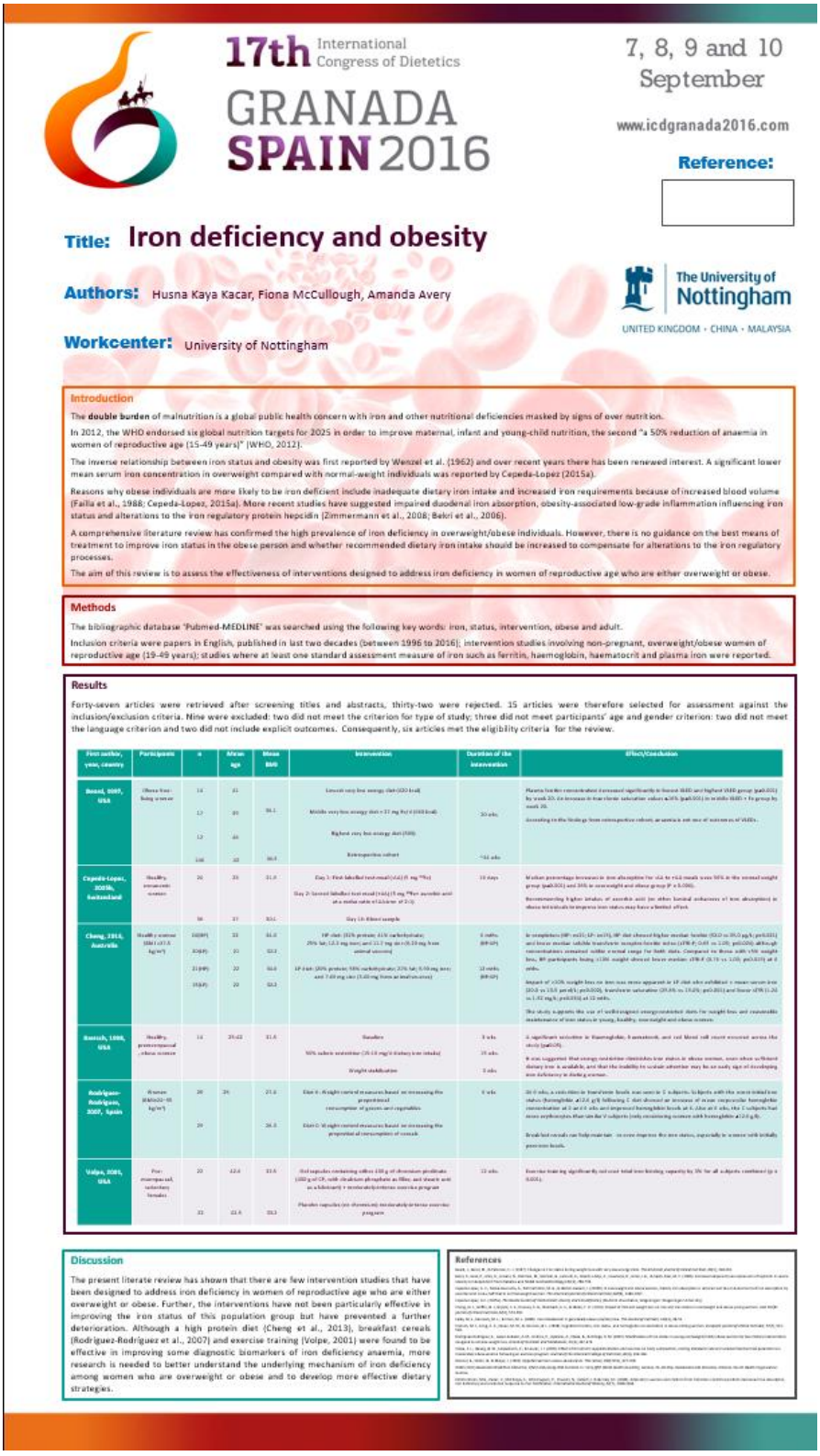
Results

The review will be completed by September 2016 and the key findings will be reported.

Conclusions, discussion and/or practical application

Obese individuals who develop iron deficiency have an increased health burden (Manios et al., 2012). The review and feasibility intervention study will inform public health recommendations in Turkey.

APPENDIX D: Poster presentation at the 17th International Congress of Dietetics, September 7-10, 2016, Granada, Spain



17th International Congress of Dietetics
GRANADA SPAIN 2016
7, 8, 9 and 10 September
www.icdgranada2016.com
Reference:

Title: Iron deficiency and obesity

Authors: Husna Kaya Kacar, Fiona McCullough, Amanda Avery

Workcenter: University of Nottingham

The University of Nottingham
UNITED KINGDOM · CHINA · MALAYSIA

Introduction

The **double burden** of malnutrition is a global public health concern with iron and other nutritional deficiencies masked by signs of over nutrition.

In 2012, the WHO endorsed six global nutrition targets for 2025 in order to improve maternal, infant and young-child nutrition, the second "a 50% reduction of anaemia in women of reproductive age (15-49 years)" (WHO, 2012).

The inverse relationship between iron status and obesity was first reported by Wenzel et al. (1962) and over recent years there has been renewed interest. A significant lower mean serum iron concentration in overweight compared with normal-weight individuals was reported by Cepeda-Lopez (2015a).

Reasons why obese individuals are more likely to be iron deficient include inadequate dietary iron intake and increased iron requirements because of increased blood volume (Failla et al., 1988; Cepeda-Lopez, 2015a). More recent studies have suggested impaired duodenal iron absorption, obesity-associated low-grade inflammation influencing iron status and alterations to the iron regulatory protein hepcidin (Zimmermann et al., 2008; Bekri et al., 2006).

A comprehensive literature review has confirmed the high prevalence of iron deficiency in overweight/obese individuals. However, there is no guidance on the best means of treatment to improve iron status in the obese person and whether recommended dietary iron intake should be increased to compensate for alterations to the iron regulatory processes.

The aim of this review is to assess the effectiveness of interventions designed to address iron deficiency in women of reproductive age who are either overweight or obese.

Methods

The bibliographic database 'Pubmed-MEDLINE' was searched using the following key words: iron, status, intervention, obese and adult.

Inclusion criteria were papers in English, published in last two decades (between 1996 to 2016); intervention studies involving non-pregnant, overweight/obese women of reproductive age (19-49 years); studies where at least one standard assessment measure of iron such as ferritin, haemoglobin, haematocrit and plasma iron were reported.

Results

Forty-seven articles were retrieved after screening titles and abstracts, thirty-two were rejected. 15 articles were therefore selected for assessment against the inclusion/exclusion criteria. Nine were excluded: two did not meet the criterion for type of study; three did not meet participants' age and gender criterion: two did not meet the language criterion and two did not include explicit outcomes. Consequently, six articles met the eligibility criteria for the review.

First author, year, country	Participants	n	Mean age	Mean BMI	Intervention	Duration of the intervention	Effect/Conclusion
Beard, 1997, USA	Obese free-living women	14 12 12 148	31 30 46 32	36.1 36.7	Lowcal very low energy diet (LCD-Lcal) Middle very low energy diet = 37 mg Fe/ d (MID-Lcal) Highest very low energy diet (VLED) Retrospective cohort	20 wks ~16 wks	Plasma ferritin concentration increased significantly in lowest (LED) and highest (VLED) group ($p<0.001$) by week 20. An increase in iron status was 36% ($p<0.001$) in middle (MED = Fe group by week 20. According to the findings from retrospective cohort, anaemia is not one of outcomes of VLEDs.
Cepeda-Lopez, 2015b, Switzerland	Healthy, premenopausal women	26 36	33 37	21.9 20.1	Day 1: First labelled test meal (+54) (5 mg ^{58}Fe) Day 2: Second labelled test meal (+162) (5 mg ^{58}Fe+ ascorbic acid at a molar ratio of 1:4/ate of 2:1) Day 14: Blood sample	14 days	Median percentage increases in iron absorption for +54 to +162 meals were 98% in the normal weight group ($p<0.001$) and 39% in overweight and obese group ($P = 0.006$). Demonstrating higher intakes of ascorbic acid (or other limited enhancers of iron absorption) in obese individuals to improve iron status may have a limited effect.
Cheng, 2014, Australia	Healthy women (BMI ≥ 27.5 kg/m²)	10(HP) 10(LP) 21(HP) 19(LP)	33 31 32 29	31.2 32.2 33.0 32.3	HP diet: (51% protein; 41% carbohydrate; 29% fat; 12.3 mg iron; and 11.7 mg dairy protein and 0.39 mg from animal sources) LP diet (20% protein; 70% carbohydrate; 21% fat; 0.56 mg iron; and 7.69 mg zinc (3.69 mg from animal sources)	6 mths (HP-LP) 12 mths (HP-LP)	In completers (HP: n=21; LP: n=19), HP diet showed higher median ferritin (52.2 vs 39.0 μg/L; p=0.021) and lower median soluble transferrin receptor-ferritin index (sTfR-F; 0.49 vs 1.09; p=0.006) although concentrations remained within normal range for both diets. Compared to those with >5% weight loss, 8% participants losing ≥5% weight showed lower median sTfR-F (0.73 vs 1.10; p=0.025) at 6 mths. Impact of >5% weight loss on iron was more apparent in LP diet who exhibited a mean serum iron (20.0 vs 13.0; p=0.01); p=0.020), transferrin saturation (29.4% vs 19.4%; p=0.001) and ferritin (79.1 vs 1.42 mg/L; p=0.016) at 12 mths. The study suggests the use of weight-support energy-restricted diets for weight loss and sustainable maintenance of iron status in young, healthy, overweight and obese women.
Beinlich, 1998, USA	Healthy, premenopausal, obese women	14	29-42	31.6	Baseline 50% calorie restriction (25-30 mg/d dietary iron intake) Weight stabilization	3 wks 10 wks 3 wks	A significant reduction in Haemoglobin, haematocrit, and red blood cell count occurred across the study ($p<0.05$). It was suggested that energy restriction diminishes iron status in obese women, even when sufficient dietary iron is available, and that the inability to sustain adherence may be an early sign of developing iron deficiency in dieting women.
Rodriguez-Rodriguez, 2007, Spain	Women (BMI=30-45 kg/m²)	20 19	34	37.8 38.3	Diet A: Weight control measures based on increasing the proportional consumption of grains and vegetables Diet O: Weight control measures based on increasing the proportional consumption of cereals	6 wks	At 0 wks, a reduction in transferrin levels was seen in 5 subjects. Subjects with the most limited iron status (haemoglobin at 12.4 g/L following 7, diet showed an increase of mean corpuscular haemoglobin concentration at 3 and 6 wks and improved haemoglobin levels at 6. Also at 6 wks, the 7 subjects had more erythrocytes than the 8 subjects (only concerning women with haemoglobin at 12.0 g/L). Breakfast cereals can help maintain – or even improve the iron status, especially in women with initially poor iron levels.
Volpe, 2001, USA	Pre-menopausal, sedentary females	20 22	22.6 21.6	33.6 33.3	Oral capsules containing either 200 μg of chromium picolinate (200 μg Cr, with chromium phosphate as filler) and stearic acid as a lubricant) + moderate/strenuous exercise program Placebo capsules (no chromium) moderately to twice exercise program	12 wks	Iron-like training significantly increased total iron-binding capacity by 5% for all subjects combined ($p \leq 0.05$).

Discussion

The present literate review has shown that there are few intervention studies that have been designed to address iron deficiency in women of reproductive age who are either overweight or obese. Further, the interventions have not been particularly effective in improving the iron status of this population group but have prevented a further deterioration. Although a high protein diet (Cheng et al., 2013), breakfast cereals (Rodriguez-Rodriguez et al., 2007) and exercise training (Volpe, 2001) were found to be effective in improving some diagnostic biomarkers of iron deficiency anaemia, more research is needed to better understand the underlying mechanism of iron deficiency among women who are overweight or obese and to develop more effective dietary strategies.

References

APPENDIX E: Abstract for poster presentation at the ASO UK Congress on Obesity 2016, September 19-20, Nottingham, UK

Poster Presentation Abstracts

28 Poster: Iron Deficiency Anaemia (IDA) and Obesity – Are the two conditions linked?

Husna Kaya Kacar[1], Fiona McCullough[1], Amanda Avery[1], Sarah-Elizabeth Bennett[2], [1]*University of Nottingham, Nottingham, UK, 2Slimming World, Derbyshire, UK*

Background: Obesity and iron deficiency are both global public health issues. A number of studies suggest that individuals who are obese are more likely to have an impaired iron status[1].

The study aim was to identify associations between body mass index (BMI) and indicators of iron status, including dietary intake, tiredness and low physical activity.

Methods: Ethical approval was obtained to carry out an online survey, with female adult members of Slimming World (SW). 533 eligible non-pregnant female participants aged 19-49 took part. The questionnaire collected data before joining SW. Data including weight, height, eating habits, past medical history (PMH) of iron deficiency anaemia, use of iron supplements and level of fatigue[2] experienced were reported using a combination of validated questionnaires. Data were analysed by BMI category.

Results: Early analysis indicates that there was no significant difference between the two BMI groups in terms of PMH of iron deficiency anaemia.

	BMI<30 kg/m²	BMI≥30 kg/m²	*p*-value
Number (%)	131 (24.6%)	365 (68.5%)	-
Mean BMI	27.2	38.0	P<0.001
Mean age	35.5	37.1	P=0.06
Diagnosis of IDA (%)	36.9%	38.6%	P=0.612
Treatment for IDA (%)	35.4%	37.9%	P=0.814
Level of fatigue (%)	38.6%	50.0%	P=0.06

Conclusion: People with a higher BMI were more likely to have experienced fatigue but the finding was not significant.

Refrerences

1. Cepeda-López, A.C. (2015). The double burden of malnutrition: obesity and iron deficiency (Doctoral dissertation, Wageningen: Wageningen University).
2. Belza, B. L., Henke, C. J., Yelin, E. H., Epstein, W. V., & Gilliss, C. L. (1993). Correlates of fatigue in older adults with rheumatoid arthritis. Nursing research, 42(2), 93-99.

APPENDIX F: Poster presentation at the ASO UK Congress on Obesity 2016, September 19-20, Nottingham, UK

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Iron deficiency and obesity
Are the two linked?

Husna Kaya Kacar[1], Sarah Bennett[2], Amanda Avery[1], Fiona McCullough[1]
1. School of Biosciences, University of Nottingham 2. Slimming World

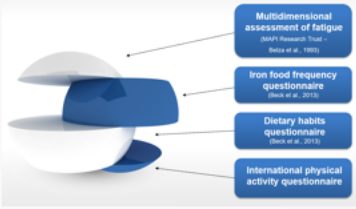
1. Introduction
- Obesity and iron deficiency are both global public health issues (WHO, 2016). A number of studies suggest that individuals who are obese are more likely to have an impaired iron status (Hassapidou *et al.*, 2006; Cepeda-Lopez *et al.*, 2010).
- The study aim was to identify associations between body mass index (BMI) and indicators of iron status, including dietary intake, tiredness and low physical activity.



2. Participants
- Adult members of Slimming World (SW), non-pregnant women of child-bearing age (19-49).

3. Methods
- Ethical approval was obtained.
- Data including weight, height, eating habits, past medical history (PMH) of iron deficiency anaemia (IDA), use of iron supplements and level of fatigue experienced were reported using a combination of validated questionnaires.
- Data were analysed by BMI category (<30 kg/m² vs. ≥30 kg/m²)

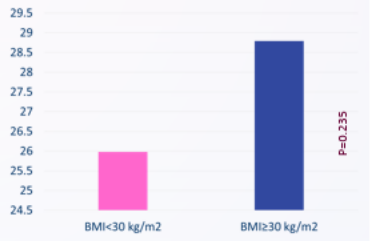
Questionnaire was developed using questions from appropriately validated tools



4. Results
- 489 eligible participants took part.
- Early analysis indicates that there was no significant difference between the two BMI groups in terms of PMH of IDA.
- Although there was a difference between two BMI group in terms of level of fatigue, the results were not statistically significant.

	BMI<30 kg/m²		BMI≥30 kg/m²		p-value
Number (%)	136		353		-
Mean age	35.3 (±8.5)	min 19 max 49	37.1 (±8.3)	min 19 max 49	p= 0.479
Mean BMI	27.25 (±1.8)	min 22.5 max 29.9	37.79 (±8.3)	min 30.02 max 57.60	p<0.001
Mean number of weeks as a SW member	2.12 (±1.6)	min 0 max 6	2.10 (±1.7)	min 0 max 7	p= 0.354
Diagnosed with IDA	Yes: 51 (37.8%)		Yes: 136 (38.5%)		p= 0.371
	No: 73 (54.1%)		No: 177 (50.1%)		
	Don't know: 11 (8.1%)		Don't know: 40 (11.3%)		
Treated for IDA	Yes: 47 (34.8%)		Yes: 134 (38.1%)		P= 0.709
	No: 82 (60.7%)		No: 208 (59.1%)		
	Don't know: 6 (4.4%)		Don't know: 10 (2.8%)		

Mean Global Fatigue Index (GFI)



5. Discussion/Conclusion
- People with a higher BMI were more likely to have experienced fatigue but the finding was not significant.
- Further analyses to be undertaken include the analysis of the iron FFQ (Beck et al., 2013) to establish whether differences in dietary iron intake might explain differences seen in IDA and GFI scores.
- Further research on the effect of weight loss programmes on fatigue might help in understanding the link between iron deficiency and obesity.

7. Further information
If you're interested in more information, please contact sbxhk1@nottingham.ac.uk

Additionally, more information about Slimming World can be found at http://www.slimmingworld.co.uk/

6. References
Beck, K. L., & Heath, A. L. M. (2013). Dietary approaches to assessing iron-related nutrition. Current Opinion in Clinical Nutrition & Metabolic Care, 16(6), 712-718.

Belza, B. L., Henke, C. J., Yelin, E. H., Epstein, W. V., & Gilliss, C. L. (1993). Correlates of fatigue in older adults with rheumatoid arthritis.Nursing research, 42(2), 93-99.

Cepeda-Lopez, A.C., Aeberli, I. & Zimmermann, M.B. (2010). Does obesity increase risk for iron deficiency? A review of the literature and the potential mechanisms. International Journal for Vitamin and Nutrition Research, 80(4), 263.

Hassapidou, M., Fotiadou, E., Maglara, E., & Papadopoulou, S. K. (2006). Energy intake, diet composition, energy expenditure, and body fatness of adolescents in northern Greece. Obesity, 14(5), 855-862.

WHO. (2016) Micronutrient deficiencies [online] Available at: http://www.who.int/nutrition/topics/ida/en/ [Accessed: 15 Feb 2015].

APPENDIX G: Abstract for poster presentation at the ASO UK

Congress on Obesity 2017, September 7-8, Pontypridd Cardiff, UK

Relationship between fatigue and level of physical activity in women who engaged in a weight management programme.

Introduction: Individuals with obesity have higher levels of fatigue than people of healthy weight. Weight management programmes encourage participants to improve both their diet and physical activity levels. This study investigates the effects of physical activity on level of fatigue in people with obesity.

Method: An online survey, hosted by Slimming World, completed by new female adult members. The questionnaire asks for before joining (T0) and current data (T1), including weight, height, level of fatigue (using the validated Multidimensional Assessment of Fatigue), and level of physical activity (IPAQ-short form). 12 weeks (T2) later, the same participants were invited to complete a second survey asking the same questions. Global fatigue index (GFI) and total Metabolic Equivalent of Task (MET) - minutes/week calculated at each time point.

Results: 74 eligible participants (BMI≥30kg/m^2) completed the questionnaire at all time-points. Early analysis indicates significant differences ($p<0.001$) for levels of physical activity and fatigue over the three time points (T0, T1 and T2) (table). For physical activity, there was a significant effect size for time, $p<.001$. The Pearson correlation coefficient suggests a very weak negative correlation between physical activity and fatigue ($r= -.092$, $n=512$, $p<.05$) at T1.

Time period	N	Mean MET-minutes/week	Standard deviation
T0	74	1383.79	1990.79
T1	74	2842.63	2982.89
T2	74	2953.17	2455.82

Table: Descriptive statistic for level of physical activity for T0, T1 and T2

Discussion: Increasing physical activity levels may help to reduce levels of fatigue in females with obesity. Further analysis will be undertaken to investigate other factors which may reduce levels of fatigue as a result of weight management programme.

APPENDIX H: Poster presentation at the ASO UK Congress on Obesity 2017, September 7-8, Pontypridd Cardiff, UK

University of Nottingham
UK | CHINA | MALAYSIA

RELATIONSHIP BETWEEN FATIGUE AND LEVEL OF PHYSICAL ACTIVITY IN WOMEN WHO ENGAGED IN A WEIGHT MANAGEMENT PROGRAMME

Husna Kaya Kacar[1], Sarah Bennett[2], Amanda Avery[1], Fiona McCullough[1]
1. School of Biosciences, University of Nottingham 2. Slimming World

1. Introduction

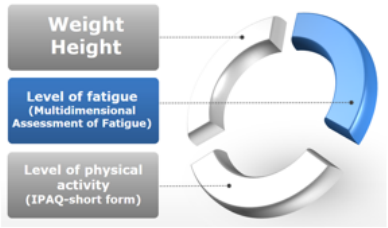
- Individuals with obesity have higher levels of fatigue than people of healthy weight [1].
- Weight management programmes encourage participants to improve both their diet and physical activity levels.
- This study investigates the effects of a weight management programme (SW) on weight loss, physical activity and level of fatigue in women with obesity.

2. Participants

- New members of Slimming World (SW); non-pregnant women of child-bearing age (19-49).

3. Method

- An online survey, hosted by Slimming World, completed by new female adult members.
- The questionnaire included before joining (T0) and current data (T1): weight, height, level of fatigue (using the validated Multidimensional Assessment of Fatigue[2], and level of physical activity (IPAQ-short form).
- 12 weeks (T2) later, the same participants were invited to complete a second survey asking the same questions. Global fatigue index (GFI) and total Metabolic Equivalent of Task (MET) - minutes/week were calculated at each time point.



4. Results

- 74 eligible participants (BMI≥30kg/m2) completed the questionnaire at all time-points.
- The analysis indicated that there were significant differences (p<0.001) over the three time points in terms of BMI, physical activity, and fatigue levels (table).
- For BMI, physical activity and fatigue, there was a significant effect size for time, p<.001.
- A significant positive correlation between weight loss and decrease in fatigue level was found (r=.27, p<.05) (figure).
- Also, physical activity level at T2 was significantly and positively correlated with weight loss (r= .24, p<.05).

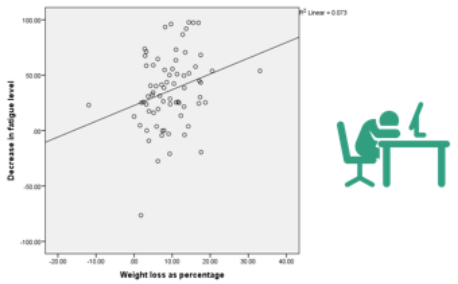


Figure: Scatter plot for the correlation between decrease in fatigue level and weight loss

	Mean (SD)			
	T0	T1	T2	P value
BMI	33.5 (±6.5)	32.38 (±6.2)	30.52 (±6.3)	<.001
PA (MET)	1383.8 (±1990.8)	2842.6 (±2982.9)	2953.2 (±2455.8)	<.001
Fatigue (GFI)	29.4 (±12.2)	20.8 (±11.6)	18.3 (±10.6)	<.001

Table: Descriptive statistics for BMI, GFI and PA for T0, T1 and T2.

5. Discussion

- Increasing physical activity levels may help to reduce levels of fatigue in females with obesity engaging in a weight management programme.
- Further analysis will be undertaken to investigate other factors which may reduce levels of fatigue.

References

1. Resnick, H. E., Carter, E. A., Aloia, M., & Phillips, B. (2006). Cross-sectional relationship of reported fatigue to obesity, diet, and physical activity: results from the third national health and nutrition examination survey. Journal of clinical sleep medicine: JCSM: official publication of the American Academy of Sleep Medicine, 2(2), 163-169.
2. Belza, B. L., Henke, C. J., Yelin, E. H., Epstein, W. V., & Gilliss, C. L. (1993). Correlates of fatigue in older adults with rheumatoid arthritis. Nursing research, 42(2), 93-99.

Slimming World
http://www.slimmingworld.co.uk

APPENDIX I: Abstract for poster presentation at the Nutrition Society Summer Conference 2018: Getting energy balance right, July 10-12, Leeds, UK

Impact of a weight management programme on the dietary habits of non-pregnant women

By H.K Kacar[1], S. Bennett[2], A. Avery[1] and F. McCullough[1] *[1]School of Biosciences, University of Nottingham, Sutton Bonington Campus, Nr Loughborough, LE12 5RD, UK, [2]Slimming World*

Weight loss maintenance and preventing weight regain leads to health improvements [(1,2)]. There is evidence that a healthy diet with fewer energy dense foods leads to weight loss and sustaining behaviour change is likely to be achieved by habit-based interventions [(3,4)]. Eating breakfast is one of the habits associated with weight loss maintenance [(5).] This study investigates the effects of a weight management programme on weight loss and changes in dietary habits.

Data was collected in the UK between May-November 2016. An online survey, hosted by Slimming World (SW), was completed by female members (19-49 years) who had recently joined the programme. The questionnaire included before joining (T0) and current data (T1): weight, height, validated food frequency and dietary habits questionnaire. 12 weeks (T2) later, the same participants were invited to complete a second survey with the same questions.

543 eligible participants completed the first questionnaire (T0 and T1), and 74 the second questionnaire (T2). Mean BMI decreased from 35.0 (±7.1) to 34.1 (±6.9) from T0 to T1 ($p<.001$) and to 31.0 (±7.2) at T2 ($p<.001$).

Data from the dietary habits questionnaire showed the percentage of participants who reported always (4 to7 times/week) having breakfast increased from T0 to T1 ($p<.001$) with no significant difference between T1 and T2 (50.8%, 83.6% and 80.6%). Whilst around two-thirds of the participants (69.4%) reported they usually ate bread or toast at breakfast at T0, this reduced to 24.5% at T1 ($p<.001$) with a small increase at T2 (35.5%). A further change at breakfast was in fruit consumption. Between T0 and T1, there was nearly a four-fold increase in fruit intake (16.8% to 65.2%, $p<.001$) with the increase at breakfast maintained at T2 (63.2%).

Bread intake at lunch-time also decreased between T0 and T1 (86.4% to 11.8%, $p=.78$) and the reduced intake was maintained at T2 (27.6%). At the evening meal, bread consumption again decreased over the three time points (33.3%, 4.2% [$p<.001$] and 3.9%). However, fruit and vegetable intake increased (13.1%, 51.0% [$p<.001$] and 27.6% [fruit]; 71.6%, 89.0% [$p<.001$] and 90.8% [veg] at T0, T1 and T2 respectively).

Investigating the changes in food intake between meals, the most significant decreases were in biscuits or cakes (79.4%, 4.4% [$p=.13$] and 27.6%), potato crisps (77.9%, 6.8% [$p=.003$] and 25.0%), and chocolate and sweets (79.6%, 6.1% [$p=.003$] and 21.1%). The most significant increases were in fruit (28.0%, 84.2% [$p=.008$] and 85.5%) and vegetables (4.6%, 44.9% [$p<.001$] and 22.4% at T0, T1, and T2 respectively).

In conclusion, this study has shown that SW is an effective weight management programme in helping to build healthy dietary habits in non-pregnant women with obesity of child-bearing age. Further analyses will determine whether any of the changes significantly contribute to the weight loss observed or whether it is a combination of the changes in dietary habits. Further research is required to investigate sustainability of these healthy habits.

1. Avenell A, Brown TJ, McGee MA *et al* (2004) *J Hum Nutr Diet* 17(4), 317-335.
2. Barte JCM, Ter Bogt NCW, Bogers RP *et al* (2010) *Obes Rev* 11(12), 899-906.
3. Crino M, Sacks G, Vandevijvere S *et al* (2015) *Curr Obes Rep* 4(1), 1-10.
4. Cleo G, Isenring E, Thomas R *et al* (2017). *J Hum Nutr Diet* 30(5), 655-664.
5. Wyatt HR, Grunwald GK, Mosca CL *et al* (2002) *Obesity* 10(2), 78-82.

APPENDIX J: Poster presentation at the Nutrition Society Summer Conference 2018: Getting energy balance right, July 10-12, Leeds, UK

Impact of a weight management programme on the dietary habits of non-pregnant women

Husna Kaya Kacar[1], Sarah Bennett[2], Amanda Avery[1], Fiona McCullough[1]
1. School of Biosciences, University of Nottingham 2. Slimming World

University of Nottingham
UK | CHINA | MALAYSIA

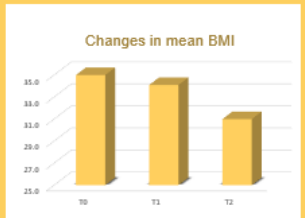


1. Introduction

- Weight loss maintenance and preventing weight regain leads to health improvements [1,2].
- There is evidence that a healthy diet with fewer energy dense foods leads to weight loss and sustaining behaviour change is likely to be achieved by habit-based interventions [3,4].
- Eating breakfast is one of the habits associated with weight loss maintenance [5].
- This study investigates the effects of a weight management programme on weight loss and changes in dietary habits.

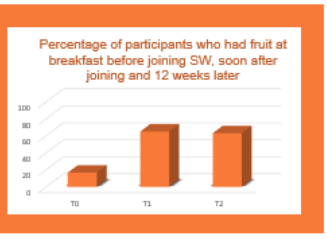
2. Methods

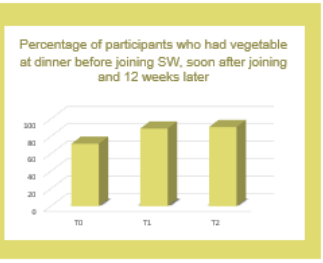
- Data was collected in the UK between May-November 2016.
- An online survey, hosted by Slimming World (SW), was completed by female members (19-49 years) who had recently joined the programme.
- The questionnaire included before joining (T0) and current data (T1): weight, height, validated food frequency and dietary habits questionnaire.
- 12 weeks (T2) later, the same participants were invited to complete a second survey with the same questions.



3. Results

- 543 eligible participants completed the first questionnaire (T0 and T1), and 74 the second questionnaire (T2).
- The percentage of participants who reported always (4 to7 times/week) having breakfast increased from T0 to T1 (p<.001) with no significant difference between T1 and T2 (50.8%, 83.6% and 80.6%).
- Around two-thirds of the participants (69.4%) reported they usually ate bread or toast at breakfast at T0. This reduced to 24.5% at T1 (p<.001) with a small increase at T2 (35.5%).
- Between T0 and T1, there was nearly a four-fold increase in fruit intake (16.8% to 65.2%, p<.001) with the increase at breakfast maintained at T2 (63.2%).
- Bread intake at lunch-time decreased between T0 and T1 (86.4% to 11.8%, p=.78) and the reduced intake was maintained at T2 (27.6%).
- At the evening meal, vegetable intake increased over the three time points (71.6%, 89.0% [p<.001] and 90.8%).
- Investigating the changes in food intake between meals, the most significant decreases were in biscuits or cakes (79.4%, 4.4% [p=.13] and 27.6%), potato crisps (77.9%, 6.8% [p=.003] and 25.0%), and chocolate and sweets (79.6%, 6.1% [p=.003] and 21.1%).





4. Conclusion

- This study has shown that SW is an effective weight management programme in helping to build healthy dietary habits in non-pregnant women with obesity of child-bearing age.
- Further analyses will determine whether any of the changes significantly contribute to the weight loss observed or whether it is a combination of the changes in dietary habits.
- Further research is required to investigate sustainability of these healthy habits.

References
1. Avenell A, Brown TJ, McGee MA *et al* (2004) *J Hum Nutr Diet* 17(4), 317-335.
2. Barte JCM, Ter Bogt NCW, Bogers RP *et al* (2010) *Obes Rev* 11(12), 899-906.
3. Crino M, Sacks G, Vandevijvere S *et al* (2015) *Curr Obes Rep* 4(1), 1-10.
4. Cleo G, Isenring E, Thomas R *et al* (2017) *J Hum Nutr Diet* 30(5), 655-664.
5. Wyatt HR, Grunwald GK, Mosca CL *et al* (2002) *Obesity* 10(2), 78-82.

APPENDIX K: Abstract for poster presentation at Women and Maternal Nutrition and Care, August 31-September 01, 2018, Toronto, Canada

Husna Kaya Kacar et al., J Nutr Food Sci 2018, Volume 8
DOI: 10.4172/2155-9600-C6-069

Annual Conference on

Women and Maternal Nutrition and Care

Aug 31-Sep 01, 2018 | Toronto, Canada

A hidden danger of obesity: Iron deficiency anaemia

Husna Kaya Kacar and **Fiona Mc Cullough**
University of Nottingham, UK

Hidden hunger is a global public health concern with iron and other nutritional deficiencies masked by signs of overnutrition. The inverse relationship between iron status and obesity was first reported in 19621. Reasons, why obesity is associated with iron deficiency, include inadequate dietary iron intake and increased iron requirements because of increased blood volume2. Impaired duodenal iron absorption, obesity-associated low-grade inflammation influencing iron status and alterations to the iron regulatory protein hepcidin have also been suggested3. The aim of the review is to evaluate interventions which treat iron deficiency anemia in obese women of reproductive age. The database 'PubMed-MEDLINE' was searched using the following keywords: iron, status, intervention, obese and adult. Intervention studies involving non-pregnant, women with overweight/obesity of reproductive age (19-49 years) and studies where ≥ one standard assessment measure of iron such as ferritin, hemoglobin, hematocrit and plasma iron were included. Forty-seven articles were assessed against the eligibility criteria and six articles were then included. The literature review has shown that few intervention studies have been designed to address iron deficiency in women of reproductive age who are either overweight or obese. Further, the interventions have not been particularly effective in improving the iron status of this population group but have prevented further deterioration. Although a high protein diet4, breakfast cereals5 and exercise training6 were found to be effective in improving some biomarkers for iron status, more research is needed to better understand the underlying mechanism of iron deficiency among women who are overweight or obese.

Biography

Kaya Kacar is currently doing Ph.D. in Nutritional Sciences at the University of Nottingham, UK. She received her bachelor's degree from the University of Erciyes, Turkey. She then worked in a public hospital in Turkey, served as a dietitian. She completed her Master in Advanced Dietetic Practice from the University of Nottingham. She is a fellow in Turkish Dietetic Association. Kaya Kaçar is awarded by the Turkish Ministry of Education Ph.D. scholarship

sbxhk1@exmail.nottingham.ac.uk

Notes:

APPENDIX L: Adjusted Downs and Black Quality Assessment Checklist

Item	Criteria	Possible Answers
Reporting		
1	*Is the hypothesis/aim/objective of the study clearly described?*	Yes = 1 No = 0
2	*Are the main outcomes to be measured clearly described in the Introduction or Methods section?* If the main outcomes are first mentioned in the Results section, the question should be answered no.	Yes = 1 No = 0
3	*Are the characteristics of the patients included in the study clearly described?* In cohort studies and trials, inclusion and/or exclusion criteria should be given. In case-control studies, a case-definition and the source for controls should be given.	Yes = 1 No = 0
4	*Are the interventions of interest clearly described?* Treatments and placebo (where relevant) that are to be compared should be clearly described.	Yes = 1 No = 0
5	*Are the distributions of principal confounders in each group of subjects to be compared clearly described?* A list of principal confounders is provided.	Yes = 2 Partially = 1 No = 0
6	*Are the main findings of the study clearly described?* Simple outcome data (including denominators and numerators) should be reported for all major findings so that the reader can check the major analyses and conclusions. (This question does not cover statistical tests which are considered below).	Yes = 1 No = 0
7	*Does the study provide estimates of the random variability in the data for the main outcomes?* In non-normally distributed data the interquartile range of results should be reported. In normally distributed data the standard error, standard deviation or confidence intervals should be reported. If the distribution of the data is not described, it must be assumed that the estimates used were appropriate and the question should be answered yes.	Yes = 1 No = 0
8	*Have all important adverse events that may be a consequence of the intervention been reported?* This should be answered yes if the study demonstrates that there was a comprehensive attempt to measure adverse events. (A list of possible adverse events is provided).	Yes = 1 No = 0
9	*Have the characteristics of patients lost to follow-up been described?* This should be answered yes where there were no losses to follow-up or where losses to follow-up were so small that findings would be unaffected by their inclusion. This should be answered no where a study does not report the number of patients lost to follow-up.	Yes = 1 No = 0
10	*Have actual probability values been reported (e.g. 0.035 rather than <0.05) for the main outcomes except where the probability value is less than 0.001?*	Yes = 1 No = 0
External validity		
11	*Were the subjects asked to participate in the study representative of the entire population from which they were recruited?* The study must identify the source population for patients and describe how the patients were selected. Patients would be representative if they comprised the entire source population, an unselected sample of consecutive patients, or a random sample. Random sampling is only feasible where a list of all members of the relevant population exists. Where a study does not report the proportion of the source population from which the patients are derived, the question should be answered as unable to determine.	Yes = 1 No = 0 Unable to determine = 0

12	*Were those subjects who were prepared to participate representative of the entire population from which they were recruited?* The proportion of those asked who agreed should be stated. Validation that the sample was representative would include demonstrating that the distribution of the main confounding factors was the same in the study sample and the source population.	Yes = 1 No = 0 Unable to determine = 0
13	*Were the staff, places, and facilities where the patients were treated, representative of the treatment the majority of patients receive?* For the question to be answered yes the study should demonstrate that the intervention was representative of that in use in the source population. The question should be answered no if, for example, the intervention was undertaken in a specialist centre unrepresentative of the hospitals most of the source population would attend.	Yes = 1 No = 0 Unable to determine = 0
Internal validity - bias		
14	*Was an attempt made to blind study subjects to the intervention they have received?* For studies where the patients would have no way of knowing which intervention they received, this should be answered yes.	Yes = 1 No = 0 Unable to determine = 0
15	*Was an attempt made to blind those measuring the main outcomes of the intervention?*	Yes = 1 No = 0 Unable to determine = 0
16	*If any of the results of the study were based on "data dredging", was this made clear?* Any analyses that had not been planned at the outset of the study should be clearly indicated. If no retrospective unplanned subgroup analyses were reported, then answer yes.	Yes = 1 No = 0 Unable to determine = 0
17	*In trials and cohort studies, do the analyses adjust for different lengths of follow-up of patients, or in case-control studies, is the time period between the intervention and outcome the same for cases and controls?* Where follow-up was the same for all study patients the answer should be yes. If different lengths of follow-up were adjusted for by, for example, survival analysis the answer should be yes. Studies where differences in follow-up are ignored should be answered no.	Yes = 1 No = 0 Unable to determine = 0
18	*Were the statistical tests used to assess the main outcomes appropriate?* The statistical techniques used must be appropriate to the data. For example nonparametric methods should be used for small sample sizes. Where little statistical analysis has been undertaken but where there is no evidence of bias, the question should be answered yes. If the distribution of the data (normal or not) is not described it must be assumed that the estimates used were appropriate and the question should be answered yes.	Yes = 1 No = 0 Unable to determine = 0
19	*Was compliance with the intervention/s reliable?* Where there was non-compliance with the allocated treatment or where there was contamination of one group, the question should be answered no. For studies where the effect of any misclassification was likely to bias any association to the null, the question should be answered yes.	Yes = 1 No = 0 Unable to determine = 0
20	*Were the main outcome measures used accurate (valid and reliable)?* For studies where the outcome measures are clearly described, the question should be answered yes. For studies which refer to other work or that demonstrates the outcome measures are accurate, the question should be answered as yes.	Yes = 1 No = 0 Unable to determine = 0
Internal validity - confounding (selection bias)		
21	*Were the patients in different intervention groups (trials and cohort studies) or were the cases and controls (case-control studies) recruited from the same population?* For example, patients for all comparison groups should be selected from the same hospital. The question should be answered unable to determine for cohort and case-control studies where there is no information	Yes = 1 No = 0 Unable to determine = 0

	concerning the source of patients included in the study.	
22	*Were study subjects in different intervention groups (trials and cohort studies) or were the cases and controls (case-control studies) recruited over the same period of time?* For a study which does not specify the time period over which patients were recruited, the question should be answered as unable to determine.	Yes = 1 No = 0 Unable to determine = 0
23	*Were study subjects randomized to intervention groups?* Studies which state that subjects were randomized should be answered yes except where method of randomization would not ensure random allocation. For example alternate allocation would score no because it is predictable.	Yes = 1 No = 0 Unable to determine = 0
24	*Was the randomized intervention assignment concealed from both patients and health care staff until recruitment was complete and irrevocable?* All non-randomized studies should be answered no. If assignment was concealed from patients but not from staff, it should be answered no.	Yes = 1 No = 0 Unable to determine = 0
25	*Was there adequate adjustment for confounding in the analyses from which the main findings were drawn?* This question should be answered no for trials if: the main conclusions of the study were based on analyses of treatment rather than intention to treat; the distribution of known confounders in the different treatment groups was not described; or the distribution of known confounders differed between the treatment groups but was not taken into account in the analyses. In non-randomized studies if the effect of the main confounders was not investigated or confounding was demonstrated but no adjustment was made in the final analyses the question should be answered as no.	Yes = 1 No = 0 Unable to determine = 0
26	*Were losses of patients to follow-up taken into account?* If the numbers of patients lost to follow-up are not reported, the question should be answered as unable to determine. If the proportion lost to follow-up was too small to affect the main findings, the question should be answered yes.	Yes = 1 No = 0 Unable to determine = 0
Power		
27*	*Did the study have sufficient power to detect a clinically important effect where the probability value for a difference being due to chance is less than 5%?* Sample sizes have been calculated to detect a difference of x% and y%.	Yes = 1 No = 0 Unable to determine = 0

*Item has been modified.

APPENDIX M: Ethical approval e-mail for study 1

S

SB-Biosciences-Ethics <SB-Biosciences-Ethics@exmail.nottingham.ac.uk>

Reply all

Mon 18/07/2016, 20:43

SB-Biosciences-Ethics; +2 more

Flag for follow up. Start by 10 October 2016. Due by 10 October 2016.

Dear Dr McCullough and Ms Kaya,

Thank you for your re-submission (Act - SB1516/38 PGR Kaya (McCullough))

This has now been through the ethical review process conducted by the School of Biosciences Research Ethics Committee (SB REC), and we are able to grant ethical approval for this project. Please note your approval code, which you may use on your forms (e.g. participant form, etc), is:

SBREC150132A

Please remember (as your research progresses) to inform the SB REC if there are any ethically relevant changes in your materials or protocols. This approval is valid for 12 months from the date of this email. The Committee also provides a further 5 year approval for any necessary work to be performed on the study which may arise in the process of publication and peer review. As appropriate, please inform any co-applicants of the outcome of this review.

We wish you all the very best for your study.

Best wishes
Kate Millar on behalf of the SB REC

...

APPENDIX N: Online survey 1 (study 1)

EXPLORING IRON INTAKE IN FEMALE SLIMMING WORLD MEMBERS

Page 1: INFORMATION ABOUT THE STUDY

Please read this information before deciding whether to take part in the study.

We're inviting you to take part in a research study conducted by the University of Nottingham. Before you decide whether to take part or not, it's important for you to understand why the research is being done and what it will involve. Please take time to read the following information carefully. If there's anything that's not clear, or if you'd like more information please contact us using the details at the end of this information sheet. Please take your time to decide whether you wish to take part in the study or not. Thank you.

What is the study about?

We're doing this research to understand more about the things that may or may not affect iron levels in our Slimming World members. To understand this we'll ask you about your eating habits, physical activity levels and tiredness levels.

Who is running the study?

Hüsna Kaya Kaçar is doing this research as part of her degree (PhD) at the University of Nottingham, supervised by Dr Fiona McCullough (Associate Professor) and Amanda Avery (Assistant Professor) and we've teamed up with Slimming World to do this research.

Why have I been chosen?

We're really keen to get the thoughts of female members who are new to Slimming World and have joined within the past 2 to 4 weeks. If you're currently pregnant, we're really sorry but you won't be able to take part in the research, as iron levels change naturally during pregnancy.

What will taking part involve?

If you decide to take part, we'd like you to complete a survey online and we'll ask for your email address so we can email you again in about three months' times to invite you to complete a second survey. Each survey will take about 30 minutes to complete.

We'll also ask you for your Slimming World membership number so that Slimming World can send us your membership information. However Slimming World won't tell us your identity when they give us this information and all your responses to the survey will be confidential.

Your responses will be anonymous, which means that we won't be able to match the responses you give with your name.

If there's any questions that you don't wish to answer, please feel free to skip these and move on to the next one.

What are the benefits of taking part?

If you take part you'll be helping us to understand more about the iron intake of women who are trying to lose weight and what factors, if any, can affect this. The findings will also help Slimming World give members the best possible support, now and in the future.

Are there any risks to taking part?

We don't expect there to be any risks to taking part but if you have any questions or experience any problems with the questionnaire, please contact Hüsna Kaya Kaçar via telephone or the email address at the end of this information sheet.

Do I have to take part?

It's up to you to decide whether or not to take part. If you do decide to take part you'll be asked to give your consent. If you do decide to take part, you are still free to withdraw at any time and without giving a reason. Taking part or not taking part will not affect your Slimming World membership in any way.

What happens if I change my mind?

That's OK, you can stop the survey at any point. We'll only look at the information in surveys that are fully completed, so if you don't finish the survey, we won't include your answers and please be assured that we'll never know your name or identity.

If you change your mind about your answers being used in the project, we can remove your survey answers before we start to do the analysis; however once we have started the analysis, we will not be able to remove them. If you do change your mind, you do not have to tell us why.

If you do change your mind please email Hüsna Kaya Kaçar from the University of Nottingham on sbxhk1@nottingham.ac.uk

What happens if something goes wrong?

We don't expect anything to go wrong, but if you have any concerns about this study, please contact Dr. Fiona McCullough (tel. 0115 951 6118, email: fiona.mccullough@nottingham.ac.uk. Address: The University of Nottingham Sutton Bonington Campus, Sutton Bonington, Leicestershire, LE12 5RD). If after speaking to Fiona, you still

don't feel that your concerns have been resolved, then please contact the School of Bioscience's Research Ethics Officer, Dr Kate Millar (tel. 0115 951 6303, email Kate.Millar@nottingham.ac.uk. Address: The University of Nottingham Sutton Bonington Campus, Sutton Bonington, Leicestershire, LE12 5RD).

What will happen to the results of the research study?

Results from the study will be used to produce a report for Hüsna's PhD project and may also be written up and published, although please be reassured you won't be identified in any reports or publications. If you would like to know the results, please contact Hüsna Kaya Kaçar from the University of Nottingham on sbxhk1@nottingham.ac.uk

Will my taking part in the study be kept confidential?

All of your information will be confidential and your questionnaires will be anonymous and given a research code by the researcher. Your data will be stored on a password protected computer accessed only by researchers from the University of Nottingham and these data will be used only in this project - we will not share any of your information with other parties. Your anonymous data will be kept for 7 years and then destroyed.

You can withdraw from the study any time before your data has been anonymized. We're really sorry but once your data has been anonymised, it will be impossible to identify your data to withdraw it.

How can I find out more?

If you'd like to know more about the survey please contact Hüsna Kaya Kaçar from the University of Nottingham on sbxhk1@nottingham.ac.uk

Thank you for considering participating in this research.

For further information please contact;

Husna Kaya Kacar

School of Biosciences, University of Nottingham

Sutton Bonington Campus

Loughborough

LEICS

LE12 5RD

Page 2: Confidentiality and consenting (agreeing) to take part in the study

Please read the following information before deciding whether or not to take part in the survey.

- I know that taking part in the study is my decision and that I can stop at any time without giving a reason
- I understand that taking part or not taking part will have no effect on my Slimming World programme or membership, and that I'm completely free to choose whether to take part or not and I do not have to say why I have read and understood the "Information about the Study" section
- I understand that if I need any more information about the study then I can contact Hüsna Kaya Kaçar from the University of Nottingham on sbxhk1@nottingham.ac.uk
- I understand that by continuing with this survey, I'm agreeing to take part in the study and to be contacted again in around 3 months' time.
- If I do not wish to be contacted again, I understand that I can email Hüsna Kaya Kaçar from the University of Nottingham on sbxhk1@nottingham.ac.uk
- I agree to provide my Slimming World membership number so that Slimming World can give my membership record information to Hüsna Kaya Kaçar from the University of Nottingham for the purposes of this research study.
- I agree to provide my email address so that Slimming World can contact me again in around three months' time to invite me to complete the second survey
- I understand that information gained from the study may be published but my name won't appear on any information I provide and everything will be kept confidential and stored securely
- I understand that if I change my mind about my answers being used in the study, I can ask for them to be removed before they are analysed by emailing Hüsna Kaya Kaçar from the University of Nottingham on sbxhk1@nottingham.ac.uk
- I understand that after my answers are analysed, Hüsna Kaya Kaçar will not be able to remove them from the study
- I understand that if I have any concerns about the research I can contact the ethics officer at the University of Nottingham on Kate.Millar@nottingham.ac.uk

By clicking on 'Next', I agree to participate in the research project.

Please just select the 'Next' button once, it may take a little while for each page to load, thank you!

Page 3: The following questions will ask for some information about you.

1. Please could you enter your Slimming World Membership number in the space below.



2. Please could you tell us how many weeks ago you joined Slimming World?

3. Email address:

Please enter a valid email address.

4. What is your date of birth?

Dates need to be in the format 'DD/MM/YYYY', for example 27/03/1980.

(dd/mm/yyyy)

5. What is your post-code?

Please enter a valid UK postcode.

6. Please tell us your height in feet and inches eg. 5ft 3inch or centimetres eg. 160.

7. Please could you tell us how much you weighed in st and lbs eg. 11st 11.4lbs or kilogram eg. 75kg just **before** you joined Slimming World?

8. Please could you tell us what your **current** weight is in st and lbs eg. 11st 11.4lbs or kilogram eg. 75kg?

8.a. If you know what your current weight is, how was it measured? (tick one option)

- Bathroom scales
- Measured on scales at Slimming World group
- Measured by my GP or at a hospital appointment
- I have guessed my weight
- Measured at pharmacy/supermarket
- Other

8.a.i. *If other, please specify*

9. Are you working at the moment?

- ○ Yes, full-time
- ○ Yes, part-time
- ○ No, I am not working

9.a. If yes, please could you tell us what your occupation is?

10. Are you currently a smoker of cigarettes? (Tick one option)

- ○ Yes
- ○ No
- ○ An ex-smoker
- ○ Prefer not to answer

10.a. If yes, roughly how many cigarettes do you smoke each day?

10.b. If an ex-smoker, how long ago did you give up smoking?

11. How would you describe your eating pattern?

- ○ Eat a variety of all foods, including animal products
- ○ Eat eggs, dairy, fish and chicken but avoid other meats
- ○ Eat eggs but avoid dairy products, all meats and fish (ovo-vegetarian)
- ○ Eat dairy products but avoid eggs, all meats and fish (lacto-vegetarian)
- ○ Eat eggs and dairy but avoid all meats and fish (ovo-lacto vegetarian)
- ○ Eat no animal products including eggs, dairy, and honey (vegan)
- ○ Eat fish or other sea food but avoid all other meats (pescetarian)
- ○ Other

11.a. *If other, please specify*

[ ]

12. Do you follow any diet for cultural or religious reasons?

- ○ Yes
- ○ No
- ○ Prefer not to answer

12.a. If yes, what type of diet do you follow?

13. Do you have or have you ever suffered from any acute or chronic illness (eg. Type 2 diabetes, cardiovascular disease, cancer)?

- Yes
- No
- Don't know

If yes, please could you tell us a little more about these?

For any of the following questions, if you don't know or can't remember, please type 'don't know' in the box provided, thank you

13.a. What is/was your diagnosis?

13.b. When was this?

13.c. Diagnosed by (eg.GP/nurse/consultant)

13.d. Any further details

14. Have you ever suffered from low iron stores, iron deficiency or iron deficiency anaemia?

- Yes
- No
- Don't know

If yes, please could you tell us a little more about these?

For any of the following questions, if you don’t know or can’t remember, please type ‘don’t know’ in the box provided, thank you

14.a. What is/was your diagnosis?

14.b. When was this?

14.c. Diagnosed by (eg.GP/nurse/consultant)

14.d. Any further details

15. Have you ever been treated for iron deficiency or iron deficiency anaemia?

- Yes
- No
- Don't know

If yes, please could you tell us a little more about these?

For any of the following questions, if you don't know or can't remember, please type 'don't know' in the box provided, thank you

15.a. Type of treatment

15.b. Duration

15.c. Any further details

16. How many full-term pregnancies have you had?

17. Please could you tell us if you had any medical condition which has resulted in blood loss?

- Yes
- No
- Don't know

17.a. If yes, please could you describe this and give approximate dates?

18. Please could you tell us if you had a blood transfusion in the last year?

- Yes
- No
- Don't know

18.a. If yes, please could you tell us why you received the transfusion?

19. Please could you tell us if you are postmenopausal?

- Yes
- No
- Prefer not to answer

19.a. If yes, please could you tell us what age your menopause started?

19.b. If no, please could you tell us how long your period typically last?

19.c. Please could you tell us how you describe your period? (Tick one option)

- Light
- Moderate
- Heavy
- Very heavy

20. Please could you tell us if you have taken any vitamin and/or mineral capsules/tablets at any time during the past year?

- Yes
- No
- Prefer not to answer

If yes, please could you tell us the brand name of the supplement, the type of supplement, the number taken and the frequency of intake and the dose (including units)?

For any of the following questions, if you don't know or can't remember, please type 'don't know' in the box provided, thank you

20.a. The brand

20.b. Type

20.c. Number per day

20.d. How many days per week

20.e. Dose

20.f. For how long

21. Please could you tell us if you have taken any other dietary supplements such as plain wheat bran (unprocessed bran, not 'All Bran', or breakfast cereal), fibre tablets, Noni juice, lecithin, evening primrose oil, performance enhancers, protein supplements, etc at any time during the past year?

- ○ Yes
- ○ No
- ○ Prefer not to answer

If yes, please could you tell us the brand name of the supplement, the type of supplement, the number taken and the frequency of intake and the dose (including units)?

For any of the following questions, if you don't know or can't remember, please type 'don't know' in the box provided, thank you

21.a. The brand

21.b. Type

21.c. Number per day

21.d. How many days per week

21.e. Dose

21.f. For how long

Thank you for completing these questions. The next set of questions will ask about your levels of fatigue, in other words your tiredness levels.

Please just select the 'Next' button once, it may take a little while for each page to load, thank you!

Page 4: These questions are about fatigue and the effect of fatigue on your activities.

For each of the following questions, please put a tick (✓) in the column headed between 1 and 10 that most closely indicates how you were feeling **before joining Slimming World (BSW)** and how you are feeling **now as a member of Slimming World (NOW)**. Please select one answer for each row.

22. To what degree did/do you experience fatigue **"before joining Slimming World" (BSW)** and **"now as a member of Slimming World" (NOW)?**

	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW											BSW
NOW											NOW

If you haven't experienced fatigue before joining Slimming World **and** now, please skip the rest of this section and move on to the next page.

For the next set of questions, if you only experienced fatigue **before** joining Slimming World, please only complete the 'BSW' boxes and leave the 'NOW' boxes blank

If you have only started experiencing fatigue **now**, after joining Slimming World, please only complete the 'NOW' boxes and leave the 'BSW' boxes blank, thank you.

23. How mild or severe was/ is the fatigue which you have been experiencing **"before joining Slimming World" (BSW)** and **"now as a member of Slimming World" (NOW)?**

	1 (Mild)	2	3	4	5	6	7	8	9	10 (Severe)	
BSW											BSW
NOW											NOW

24. To what degree did/does fatigue cause you distress **"before joining Slimming World" (BSW)** and **"now as a member of Slimming World" (NOW)?**

	1 (No distress)	2	3	4	5	6	7	8	9	10 (A great deal of distress)	
BSW											BSW
NOW											NOW

For each of the following questions, please put a tick (✓) in the column headed between 1 and 10 that most closely indicates to what degree fatigue has interfered with your ability to do the following activities **"before joining Slimming World" (BSW)** and **"now as a member of Slimming World" (NOW).** For activities you didn't/don't do, for reasons other than fatigue (e.g. you don't work because you are retired), **please put a tick (✓) in the column headed N/A (not applicable).** Please select one answer for each row.

25. To what degree has fatigue interfered with your ability to do household chores

	N/A	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW												BSW
NOW												NOW

26. To what degree has fatigue interfered with your ability to cook

	N/A	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW												BSW
NOW												NOW

27. To what degree has fatigue interfered with your ability to bathe or wash

	N/A	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW												BSW
NOW												NOW

28. To what degree has fatigue interfered with your ability to dress

	N/A	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW												BSW
NOW												NOW

29. To what degree has fatigue interfered with your ability to work

	N/A	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW												BSW
NOW												NOW

30. To what degree has fatigue interfered with your ability to visit or socialise with friends or familiy

	N/A	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW												BSW
NOW												NOW

31. To what degree has fatigue interfered with your ability to engage in sexual activity

	N/A	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW												BSW
NOW												NOW

32. To what degree has fatigue interfered with your ability to engage in leisure and recreational activities

	N/A	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW												BSW
NOW												NOW

33. To what degree has fatigue interfered with your ability to shop and do errands

	N/A	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW												BSW
NOW												NOW

34. To what degree has fatigue interfered with your ability to walk

	N/A	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW												BSW
NOW												NOW

35. To what degree has fatigue interfered with your ability to exercise, other than walking

	N/A	1 (Not at all)	2	3	4	5	6	7	8	9	10 (A great deal)	
BSW												BSW
NOW												NOW

36. Please could you tell us how often you felt/feel fatigued during an entire week before joining Slimming World (BSW) and over the past week as a member of Slimming World (NOW)? (Please tick one box for BSW and one box for NOW.)

	BSW	NOW
Everyday	☐	☐
Mostly, but not all days	☐	☐
Occasionally, but not most days	☐	☐
Hardly any days	☐	☐

Please just select the 'Next' button once, it may take a little while for each page to load, thank you!

Page 5: The questions below ask about the different foods you ate before joining Slimming World and those you eat now as a member of Slimming World. You might find they take a little while to complete but these questions are really important to help us to understand more about your iron intake.

For each food there are two lines **“BSW”** and **“NOW”**. Please put a tick (✓) on the **"BSW"** line in the box to indicate how often, on average, you ate the specified amount of each food before joining Slimming World. Then please put a tick (✓) on the **"NOW"** line in the box to indicate how often, on average, you now eat the specified amount of each food as a member of Slimming World. **Please tick one box on each line.** For each food there is an amount shown, either a “medium serving” or a common household unit such as a slice or teaspoon.

If you are vegetarian or vegan, please just complete the sections relevant to you.

MEAT, FISH AND CHICKEN

How often did/do you eat the following foods?

37. Beef (eg. roast, steak, chops, silverside, casseroles, stir fry, curry, mince dishes) [the size of a deck of playing cards (60–90g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

38. Chicken, turkey or duck (eg. roast, fried, steamed, BBQ, casseroles, stir fry, curry, fried takeaway chicken) [the size of a deck of playing cards (60–90g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

39. Lamb, mutton (eg. roast, steak, chops, BBQ, casseroles, stir fry, curry) [the size of a deck of playing cards (60–90g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

40. Pork (eg. roast, chops, steak, casserole, casseroles, stir fry, curry) [the size of a deck of playing cards (60–90g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

41. Veal [in breadcrumb, fried (150g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

42. Liver, kidney, other offal (including pate) [1 slice lamb's liver (40g), 1 tablespoon cooked kidney (40g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

43. Ham, bacon [1 average slice ham (23g), 1 rasher back bacon (25g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

44. Game meats (eg. venison, mutton bird, rabbit) [the size of a chequebook (120g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

45. Corn beef, canned [1 small stewed canned (200g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

PREPARED MEAT

How often did/do you eat the following foods?

46. Cured beef [1 slice 25cm x 19cm x 0.2cm (50g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

47. Sausages, frankfurters, saveloys [1 medium sausage roll (60g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

48. Luncheon sausage, salami, brawn, pastrami [1 snack salami (peperami) (25g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

49. Black pudding [1 slice (30g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

50. Meat pies [1 pasty 14cm x 8.5cm x 3.5cm (160g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

SEAFOOD

How often did/do you eat the following foods?

51. Fresh and frozen fish (eg. cod, haddock, flounder, hoki, salmon, white bait, shark, eel) [the size of a chequebook (120g)

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

52. Battered and crumbed fish (eg. fish fingers, fish cakes) [1 fish finger (28g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

53. Canned and bottled fish (eg. tuna, salmon, herrings, sardines) [1 small can tuna (100g), 1 canned sardine (25g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

54. Mussels, cockles, oysters [1 small jar mussels (80g), 1 dozen oysters (120g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

55. Scallops, crab sticks, crab, squid, crayfish [1 tablespoon crab meat (40g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

56. Prawns, shrimps [1 small jar prawns (100g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

FRUIT

How often did/do you eat the following foods?

57. Apples [1 medium (112g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

58. Bananas, green bananas [1 medium (100g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

59. Citrus fruits (eg. orange, tangerine, mandarin, grapefruit, lemon) [1 medium orange (160g), 1 slice lemon (20g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

60. Kiwifruit [1 medium (60g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

61. Pears [1 medium (150g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

62. Stone fruit (eg. apricots, nectarines, peaches, plums, lychees) [1 medium peach (110g), 1 medium plum (55g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

63. Avocadoes, olives [1 medium avocado (145g), 5 olives (15g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

64. Grapes [1 small bunch (100g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

65. Mango [1 mango (150g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

66. Watermelon [1 cup, diced (152g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

67. Pawpaw (papaya), other melons (eg. honey dew, rock melon) [1 average slice (140g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

68. Pineapple [1 large slice (80g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

69. Rhubarb [stewed with sugar, 1 cup (240g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

70. Fruit salad, canned [1 cup (263g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

71. Strawberries, blackberries, cherries, blueberries, boysenberries, loganberries, cranberries, gooseberries, raspberries [½ cup (82g), 10 cherries (67g), 10 strawberries (56g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

72. Sultanas, raisins, currants, figs [1 tablespoon, dried sultanas (30g), 3 dried figs (48g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

73. Dried apricots, prunes, dates, mixed dried fruit [5 Prunes (42g), 10 apricot halves (35g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

VEGETABLES

How often did/do you eat the following foods?

74. Potato (eg. boiled, mashed, baked, roasted, fried, chips) [1 potato (90g), 1 tablespoon mashed (45g), 1 cup 6cm x 1cm x 1cm chips (95g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

75. Sweet potato (eg. boiled, mashed, baked, roasted, fried, chips) [½ cup mashed (120g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

76. Green beans, broad beans, runner beans, asparagus [½ cup boiled green bean (70g), 5 spears asparagus (125g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

77. Broccoli (all varieties) [½ cup cooked, drained, chopped (78g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

78. Red cabbage [½ cup boiled, drained, shredded (75g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

79. Cabbage (all varieties), brussel sprouts [5 brussels sprouts (100g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

80. Peppers (all varieties) [1 medium green or red pepper (160g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

81. Carrots [1 medium boiled carrot (80g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

82. Cauliflower [1 stem and flower (90g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

83. Corn (all varieties) [1 tablespoon (30g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

84. Courgette, zucchini, cucumber, gerkins or marrow (all varieties) [1 medium cooked courgette (100g), 1 inch piece cucumber (60g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

85. Beetroot, radishes (all varieties) [1 small whole beetroot (35g), ½ cup raw radishes, slices (58g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

86. Lettuce [1 cup shredded iceberg (80g)]

	Never or	1 to 3		2 to 3	4 to 6		2 to 3	4 plus	

	less than once per month	times per month	Once per week	times per week	times per week	Once per day	times per day	times per day	
BSW									BSW
NOW									NOW

87. Mushrooms [½ cup cooked (72g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

88. Onions (all varieties), leeks, celery [1 small onion (60g); 1 medium, boiled leek (160g); ½ cup celery, boiled, chopped (60g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

89. Tomatoes (all varieties) [1 medium tomato (85g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

90. Peas, green [½ cup canned, drained (85g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

91. Spinach, silver beet, swiss chard (all varieties) [½ cup boiled, chopped (75g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

92. Other green leafy vegetables (eg. watercress, chicory, kale, chard, collards, Chinese kale) [½ cup boiled, drained (73g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

93. Pumpkin, squash, yams [½ cup yam (110g),½ cup mashed pumpkin (120g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

94. Parsnip [½ cup boiled, chopped (75g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

LEGUMES

How often did/do you eat the following foods?

95. Tofu, soybeans, tempeh [1 cup tofu (262g) or 1 cup soybeans, cooked (94g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

96. Beans in sauce (eg. baked beans, chilli beans) [½ cup (94g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

97. Beans (canned or dried) (eg. black beans, butter beans, haricot beans, red kidney beans, white kidney beans, refried beans) [½ cup canned (95g)]

	Never or	1 to 3		2 to 3	4 to 6		2 to 3	4 plus	

	less than once per month	times per month	Once per week	times per week	times per week	Once per day	times per day	times per day	
BSW									BSW
NOW									NOW

98. Lentils (eg. dahl) [½ cup dried, boiled (95g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

99. Peas (eg. chick peas, hummus, falafels, split peas) [½ cup chickpea, canned, drained (95g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

BREAKFAST CEREAL

How often did/do you eat the following foods?

100. Porridge, rolled oats, oat bran, oat meal [1 cup (260g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

101. Muesli (all varieties) [1 cup (107g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

102. Weetabix (all varieties) [1 biscuits (20g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

103. Cornflakes or rice krispies [1 cup (31g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

104. Bran based cereals (all varieties eg. All Bran, Sultana Bran) [1 cup (60g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

105. Light and fruity cereals (eg. Special K, Light and tasty) [1 cup (60g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

106. Chocolate based cereals (eg. CocoPops) [1 cup (40g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

BREADS, CAKES, BISCUITS AND CRACKERS

How often did/do you eat the following foods?

108. White bread and rolls (including specialty breads such as foccacia, panini, pita, naan, crumpets, pizza bases, tortilla's, burrito, roti) [Small loaf, 1 slice (25g)

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

109. Wholegrain bread and rolls (including multigrain, wholegrain, whole meal breads) [Small loaf, 1 slice (25g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

110. Fruit and currant bread/buns [a medium bun (80g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

111. White flour muffins (all varieties) [1 muffin (80g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

112. Cakes (all varieties excluding chocolate and fruit cake) [1 slice (100g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

113. Chocolate cake [1 slice (108g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

114. Fruit cake [1 slice (40g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

115. Biscuits, plain sweet [2 biscuits (41g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

116. Biscuits, chocolate or chocolate covered [2 biscuits (34g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

117. Crackers (eg. crisp bread, water crackers, rice cakes, cream crackers) [3 crackers (27-30g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

GRAINS

How often did/do you eat the following foods?

118. White rice [1 cup, cooked (190g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

119. Brown rice [1 cup, cooked (195g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

120. Instant noodles [1 cup, cooked (160g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

121. Pasta, noodles (white) [1 cup macaroni, boiled (180g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

122. Pasta, noodles (whole wheat) [1 cup spaghetti, boiled (170g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

123. Couscous, polenta [1 cup couscous, cooked (157g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

124. Bulgur wheat (eg. tabbouleh) [1 cup, cooked (182g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

125. Wheat germ, wheat bran (flakes) [1 cup (115g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

DAIRY PRODUCTS

How often did/do you eat/drink the following foods or drinks?

126. Hard cheese (eg. Cheddar, Red Leicester, Edam, Tasty, blue vein, camembert, parmesan, gouda, processed) [the size of a small matchbox (30g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

127. Cottage cheese, ricotta cheese [1 tablespoon (40g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

128. Cream, sour cream, cream cheese, cheese spreads, fromage frais (all varieties) [1 Tablespoon (25g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

129. Milk (cow's milk) **as a drink** (eg. flavored milk, milk shakes) [1 cup (265g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

130. Milk (cow's milk) (all varieties) **added to drinks** (eg. in tea, coffee) [30-45ml]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

131. Milk (cow's milk) (all varieties) **added to food** (eg. cereals, dishes such as macaroni cheese, milk puddings such as rice pudding, custard, semolina, instant puddings, dairy food) [125ml]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

132. Soy Milk [1 cup (265g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

133. Coconut milk [1 cup (265g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

134. Yoghurt [1 cup (245g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

135. Ice cream [1 average scoop 60g]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

EGGS

How often did/do you eat the following foods?

136. Eggs – boiled, fried, poached, scrambled, raw and egg based dishes including quiche, soufflés, frittatas, omelettes [1 egg 49g]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

NUTS

How often did/do you eat the following foods?

137. Peanuts, mixed nuts, macadamias, pecan, hazelnuts, brazil nuts, walnuts, cashews, pistachios [10 whole peanuts (13g), 1 pecan (6g), 10 cashew (10g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

138. Almonds [6 whole (13g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

139. Pumpkin seeds, sunflower seeds, pinenuts [1 tablespoon sunflower seed (16g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

140. Sesame seeds, tahini [1 heaped teaspoon (19g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

MISCELLANEOUS FOODS AND SNACKS

How often did/do you eat the following foods?

141. Marmite [1 level teaspoon (9g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

142. Chocolate spread (eg. Nutella) [1 tablespoon (14g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

143. Peanut butter [1 tablespoon (15g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

144. Butter or margarine [1 tablespoon (15g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

145. Cooking oil (all varieties) [1 tablespoon (11g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

146. Soup, vegetable based, homemade or canned [1 cup (257g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

147. Sugar (all varieties) [1 teaspoon (4g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

148. Soup, meat based, homemade or canned [1 cup (253g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

149. Muesli bar, fruit based [1 bar (32g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

150. Muesli bar, nut based [1 bar (47g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

151. Muesli bar, chocolate chip or chocolate coated [1 bar (32g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

152. Potato crisps [1 small packet (50g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

153. Milk chocolate [1 small bar (50g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

154. Dark chocolate [1 small bar (50g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

155. White chocolate [1 small bar (50g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

ALCOHOL

How often did/do you drink the following drinks?

156. Beer, cider (all varieties) [1 can (335ml)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

157. Red wine [1 small glass (125ml)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

158. White wine [1 small glass (125ml)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

159. Spirits (all varieties) [1 measure (25ml)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

160. Ready to drink alcoholic beverages (eg. Alcopops or cans of pre-mixed drink)

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

NON-ALCOHOLIC BEVERAGES

How often did/do you drink the following drinks?

161. Complan, Ensure (all varieties)

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

162. Hot chocolate, drinking chocolate, Cocoa, Ovaltine, Nesquik [2 teaspoon (6g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

163. Coffee (all varieties) [1 teaspoon (2g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

164. Black tea [1 cup (250ml)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

165. Herbal tea, fruit tea [1 cup (250ml)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

166. Cordials (including syrups, powders) (eg. blackcurrant, orange) [1 Tablespoon (19g)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

167. Fruit and vegetable juices (all varieties) [1 cup fruit juice (256ml), vege juice (200ml)]

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

168. Sports drinks (eg. Powerade)

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

169. Energy drinks (eg. Red Bull)

	Never or less than once per month	1 to 3 times per month	Once per week	2 to 3 times per week	4 to 6 times per week	Once per day	2 to 3 times per day	4 plus times per day	
BSW									BSW
NOW									NOW

Thank you for completing these questions. The next set of questions will ask more detailed questions about your dietary habits.

Please just select the 'Next' button once, it may take a little while for each page to load, thank you!

Page 6: The next set of questions will ask about your dietary habits.

When answering the questions below please consider your intake of food before joining Slimming World and now as a member of Slimming World. For each time periods/foods there are two lines as **"BSW"** and **"NOW"**. Please put a tick (✓) on BSW line for before joining Slimming World and put a tick (✓) on NOW for now as a member of Slimming World.

170. At which of the following time periods did/do you eat or drink usually? Please tick one box on each line. (We realise that everyone has different eating patterns. Feel free to decide for yourself which eating occasion you choose to call breakfast, etc no matter what time it happens in the day.)

	Always (4 to 7 times per week)	Sometimes (2 to 3 times per week)	Never (0 to 1 times per week)	
Before breakfast - BSW				BSW
Before breakfast - NOW				NOW
Breakfast - BSW				BSW
Breakfast - NOW				NOW
Between breakfast and lunch - BSW				BSW
Between breakfast and lunch - NOW				NOW

Lunch - BSW				BSW
Lunch - NOW				NOW
Between lunch and evening meal - BSW				BSW
Between lunch and evening meal - NOW				NOW
Evening meal - BSW				BSW
Evening meal - NOW				NOW
After evening meal - BSW				BSW
After evening meal - NOW				NOW
Other eg. midnight snack - BSW				BSW
Other eg. midnight snack - NOW				NOW

171. If you ate/eat breakfast what types of food did/do you usually eat?

	BSW	NOW
Breakfast cereals		
Porridge		
Bread or toast		
Noodles or rice		
Milk products (eg. milk on cereal, yoghurt, dairy food, cheese)		
Fruit		
Meats (eg. bacon, sausages)		
Baked beans or eggs		

Vegetables		

172. Did/Do you usually have a drink up to 1 hour before, with, or up to 1 hour after your breakfast? Please tick one box for BSW and one box for NOW.

	BSW	NOW
YES	☐	☐
NO	☐	☐

173. If yes, what drinks did/do you usually have with your breakfast? (tick as many as apply)

	BSW	NOW
Fruit or vegetable juices	☐	☐
Milk or milk-based drinks (all varieties)	☐	☐
Soy-based drinks	☐	☐
Chocolate-based drinks (eg. drinking chocolate)	☐	☐
Coffee (all varieties)	☐	☐
Tea (all varieties)	☐	☐
Water (all varieties)	☐	☐
Other	☐	☐

174. If you ate/eat lunch, what types of food did/do you usually eat? (tick as many as apply)

	BSW	NOW
Bread (eg. sandwiches, rolls)	☐	☐
Starchy food (eg. pasta, rice, potato)	☐	☐
Milk products (eg. yoghurt, dairy food, cheese)	☐	☐
Fruit	☐	☐
Meats (eg. meat, fish, chicken, seafood, ham, salami)	☐	☐
Legumes, nuts, eggs	☐	☐
Vegetables (including salad)	☐	☐

175. Did/Do you usually have a drink up to 1 hour before, with, or up to 1 hour after your lunch? Please tick one box for BSW and one box for NOW.

	BSW	NOW
YES	☐	☐
NO	☐	☐

176. If yes, what drinks did/do you usually have with your lunch? (tick as many as apply)

	BSW	NOW
Fruit or vegetable juices	☐	☐
Milk or milk-based drinks (all varieties)	☐	☐
Soy-based drinks	☐	☐
Chocolate-based drinks (eg. drinking chocolate)	☐	☐
Coffee (all varieties)	☐	☐
Tea (all varieties)	☐	☐
Water (all varieties)	☐	☐
Other	☐	☐

177. If you ate/eat an evening meal, what types of food did/do you usually eat? (tick as many as apply)

	BSW	NOW
Bread (eg. sandwiches, rolls, etc)	☐	☐
Starchy food (eg. pasta, rice, potato)	☐	☐
Milk products (eg. yoghurt, dairy food, cheese, custard, ice cream)	☐	☐
Meats (eg. meat, fish, chicken, seafood, ham)	☐	☐
Legumes, nuts, eggs	☐	☐
Vegetables (including salad)	☐	☐
Fruit	☐	☐

178. Did/Do you usually have a drink up to 1 hour before, with, or up to 1 hour after your evening meal? Please tick one box for BSW and one box for NOW.

	BSW	NOW
YES	☐	☐
NO	☐	☐

179. If yes, what drinks did/do you usually have with your evening meal? (tick as many as apply)

	BSW	NOW
Fruit or vegetable juices	☐	☐
Milk or milk-based drinks (all varieties)	☐	☐
Soy-based drinks	☐	☐
Chocolate-based drinks (eg. drinking chocolate	☐	☐
Coffee (all varieties)	☐	☐
Tea (all varieties)	☐	☐
Alcohol	☐	☐
Water (all varieties)	☐	☐
Other	☐	☐

180. If you ate/eat or drink between meals, what types of food or drink did/do you usually have? (tick as many as apply)

	BSW	NOW
Bread based snacks (eg. sandwiches)	☐	☐
Breakfast cereal	☐	☐
Porridge	☐	☐
Biscuits or cakes	☐	☐
Crackers	☐	☐
Milk products (eg. yoghurt, dairy food, cheese, ice cream)	☐	☐
Fruit	☐	☐
Potato crisps	☐	☐
Cereal bars, muesli bars	☐	☐
Chocolate or sweets	☐	☐
Vegetables	☐	☐
Fruit or vegetable juices	☐	☐
Milk or milk-based drinks (all varieties)	☐	☐
Soy-based drinks	☐	☐
Chocolate-based drinks (eg. drinking chocolate)	☐	☐
Coffee (all varieties)	☐	☐
Tea (all varieties)	☐	☐
Water (all varieties)	☐	☐
Other	☐	☐

181. If you ate/eat or drink after your evening meal what types of food or drink did/do you usually have? (tick as many as apply)

Bread based snacks (eg. sandwiches)	BSW	NOW
Breakfast cereal	☐	☐
Porridge	☐	☐
Biscuits or cakes	☐	☐
Crackers	☐	☐
Milk products (eg. yoghurt, dairy food, cheese, ice cream)	☐	☐
Fruit	☐	☐
Potato crisps	☐	☐
Cereal bars, muesli bars	☐	☐
Chocolate or sweets	☐	☐
Vegetables	☐	☐
Fruit or vegetable juices	☐	☐
Milk or milk-based drinks (all varieties)	☐	☐
Soy-based drinks	☐	☐
Chocolate-based drinks (eg. drinking chocolate	☐	☐
Coffee (all varieties)	☐	☐
Tea (all varieties)	☐	☐
Alcohol	☐	☐
Water (all varieties)	☐	☐
Other	☐	☐

182. During the entire week (including weekdays and weekends) how many times did/do you eat out (for example, at a restaurant, food court or a café)? Please tick one box for BSW and one box for NOW.

	BSW	NOW
Never	☐	☐
Once	☐	☐
Twice	☐	☐
Three times	☐	☐
4 to 5 times	☐	☐
6 to 7 times	☐	☐
8 to 9 times	☐	☐
10 to 11 times	☐	☐
12 to 13 times	☐	☐
14 plus times	☐	☐

We appreciate that you have already told us quite a lot of detail about your food intake. These following questions just provide us with some helpful extra information. The questions will ask separately about your eating habits during the week and at the weekend.

How many servings of fruit did/do you usually eat per day (one serving = one apple, one banana, one orange, two small apricots or plums, half a cup of fruit salad or stewed fruit, one cup of juice, two tablespoons of raisins or three dates) **before joining Slimming World (BSW)** and **now as a member of Slimming World (NOW)**?

184. BSW - Number of servings per day during the week (Monday to Friday)

185. BSW - Number of servings per day at the weekend (Saturday and Sunday)

186. NOW - Number of servings per day during the week (Monday to Friday)

187. NOW - Number of servings per day at the weekend (Saturday and Sunday)

How many servings of vegetables did/do you usually eat per day (one serving = one medium potato or sweet potato, one tomato, half a cup of cooked vegetables or salad vegetables) **before joining Slimming World (BSW)** and **now as a member of Slimming World (NOW)?**

188. BSW - Number of servings per day during the week (Monday to Friday)

189. BSW - Number of servings per day at the weekend (Saturday and Sunday)

190. NOW - Number of servings per day during the week (Monday to Friday)

191. NOW - Number of servings per day at the weekend (Saturday and Sunday)

How many servings of meat, fish, chicken or seafood did/do you usually eat per day (one serving = two slices of cooked meat, one medium steak, ¾ cup of mince or casserole, one medium fillet of fish, two chicken drumsticks or one chicken leg) **before joining Slimming World (BSW)** and **now as a member of Slimming World (NOW)?**

192. BSW - Number of servings per day during the week (Monday to Friday)

193. BSW - Number of servings per day at the weekend (Saturday and Sunday)

194. NOW - Number of servings per day during the week (Monday to Friday)

195. NOW - Number of servings per day at the weekend (Saturday and Sunday)

196. Some foods and drinks have iron added to them (eg. some breakfast cereals). When you were/are choosing foods and drinks, how often did/do you choose the product with added iron instead of the product without? Please tick one box for BSW and one box for NOW.

	BSW	NOW
Whenever I can	☐	☐
Usually	☐	☐
Sometimes	☐	☐
Never	☐	☐
I don't know or I don't consider whether foods have iron added to them	☐	☐

197. How often did/do you use a cast-iron fry pan, wok or pot when preparing your meals? Please tick one box for BSW and one box for NOW.

	BSW	NOW
Never or less than once a month	☐	☐
1 to 3 times per month	☐	☐
Once per week	☐	☐
2 to 3 times per week	☐	☐
4 to 6 times per week	☐	☐
Once per day	☐	☐
2 to 3 times per day	☐	☐
4 plus times per day	☐	☐
I don't know	☐	☐
	☐	☐

198. Did/Do your meals differ at the weekend (Saturday and Sunday) compared with during the week (Monday to Friday)? Please tick one box for BSW and one box for NOW.

	BSW	NOW
Always	☐	☐
Usually	☐	☐
Sometimes	☐	☐
Never		

198.a. If sometimes, usually or always please comment.

Thank you for completing these questions. You're almost finished! The final section asks a few questions about your physical activity levels.

Please just select the 'Next' button once, it may take a little while for each page to load, thank you!

Page 7: 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 before joining Slimming World (BSW) and in the last 7 days (NOW). Please answer each question even if you do not consider yourself to be an active person. Please think about the activities you did/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 **before joining Slimming World (BSW)** and in the **last 7 days (NOW).**
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.

Before joining Slimming World (BSW) and during the last 7 days (NOW), on how many days did you do **vigorous** physical activities like heavy lifting, digging, aerobics, or fast bicycling during the entire week?

If you did/do no vigorous physical activities, please enter '0' into the box

199. BSW - How many days per week

200. NOW - How many days per week

Before joining Slimming World (BSW) and during the last 7 days (NOW), how much time did you usually spend doing vigorous physical activities on one of those days?

If you're not sure or can't remember, please enter 'not sure' in the box

201. BSW - How many hours per day

202. BSW - How many minutes per day

203. NOW – How many hours per day

204. NOW- How many minutes per day

Think about all the **moderate** activities that you did **before joining Slimming World (BSW)** and in **the last 7 days (NOW).**
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.

Before joining Slimming World (BSW) and during the last 7 days (NOW), on how many days did you do **moderate** physical activities like carrying light loads, bicycling at a regular pace, or doubles tennis during the entire week? Do not include walking.

If you did/do no moderate physical activities, please enter '0' into the box

205. BSW - How many days per week

206. NOW - How many days per week

Before joining Slimming World (BSW) and during the last 7 days (NOW), how much time did you usually spend doing **moderate** physical activities on one of those days?

If you're not sure or can't remember, please enter 'not sure' in the box

207. BSW - How many days per week

208. NOW - How many days per week

Before joining Slimming World (BSW) and during the last 7 days (NOW), how much time did you usually spend doing **moderate** physical activities on one of those days?

If you're not sure or can't remember, please enter 'not sure' in the box

207. BSW - How many hours per day

208. BSW - How many minutes per day

209. NOW - How many hours per day

210. NOW - How many minutes per day

Think about the time you spent **walking before joining Slimming World (BSW)** and **in the last 7 days (NOW).** This includes at work and at home, walking to travel from place to place, and any other walking that you have done solely for recreation, sport, exercise, or leisure.

Before joining Slimming World (BSW) and during the last 7 days (NOW), on how many days did you **walk** for at least 10 minutes at a time during the entire week?

If you did/do no walking, please enter '0' into the box

211. BSW - How many days per week

212. NOW - How many days per week

Before joining Slimming World (BSW) and during the last 7 days (NOW), how much time did you spend **walking** on one of those days?

If you're not sure or can't remember, please enter 'not sure' in the box

213. BSW - How many hours per day

214. BSW - How many minutes per day

215. NOW - How many hours per day

216. NOW - How many minutes per day

The last question is about the time you spent **sitting** on weekdays **before joining Slimming World (BSW)** and during **the last 7 days (NOW).** 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.

Before joining Slimming World (BSW) and during the last 7 days (NOW), how much time did you spend **sitting** on a week day?

If you're not sure or can't remember, please enter 'not sure' in the box

217. BSW - How many hours per day

218. BSW - How many minutes per day

219. NOW - How many hours per day

220. NOW - How many minutes per day

Please just select the 'Next' button once, it may take a little while for each page to load, thank you!

Page 8: You've now reached the end of the questionnaire and we're really grateful to you for completing this. Thank you so much! As we mentioned at the start of the survey, we'll email you again in around 3 months' time to give you a link to the second survey. If you'd rather not complete another survey in 3 months' time then please let us know by emailing Husna Kaya Kacar from University of Nottingham on sbxhk1@nottingham.ac.uk Thanks again

APPENDIX O: Total variance explained table (T0)

Total Variance Explained

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings			Rotation Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	4.254	12.154	12.154	4.254	12.154	12.154	2.861	8.173	8.173
2	3.739	10.682	22.837	3.739	10.682	22.837	2.803	8.008	16.181
3	2.152	6.149	28.985	2.152	6.149	28.985	2.320	6.629	22.811
4	1.710	4.885	33.870	1.710	4.885	33.870	2.244	6.411	29.222
5	1.691	4.831	38.701	1.691	4.831	38.701	1.921	5.489	34.710
6	1.356	3.874	42.575	1.356	3.874	42.575	1.696	4.845	39.556
7	1.321	3.776	46.351	1.321	3.776	46.351	1.539	4.397	43.952
8	1.189	3.397	49.747	1.189	3.397	49.747	1.504	4.298	48.250
9	1.169	3.339	53.086	1.169	3.339	53.086	1.364	3.898	52.148
10	1.062	3.033	56.119	1.062	3.033	56.119	1.242	3.549	55.697
11	1.010	2.886	59.004	1.010	2.886	59.004	1.158	3.307	59.004
12	.993	2.837	61.841						
13	.919	2.624	64.466						
14	.885	2.528	66.994						
15	.833	2.379	69.373						
16	.804	2.296	71.669						
17	.792	2.263	73.932						
18	.758	2.165	76.096						
19	.735	2.101	78.198						
20	.678	1.936	80.134						
21	.649	1.853	81.987						
22	.624	1.782	83.769						
23	.598	1.708	85.477						
24	.561	1.604	87.081						
25	.553	1.580	88.661						
26	.522	1.493	90.153						
27	.494	1.410	91.563						
28	.462	1.319	92.882						
29	.442	1.264	94.146						
30	.429	1.227	95.372						
31	.386	1.102	96.474						
32	.369	1.054	97.528						
33	.335	.957	98.485						
34	.301	.860	99.345						
35	.229	.655	100.000						

Extraction Method: Principal Component Analysis.

APPENDIX P: Result of Monte Carlo PCA for parallel analysis (T0)

```
11/03/2017   17:50:45
Number of variables:     35
Number of subjects:     543
Number of replications: 100

+++++++++++++++++++++++++++++++++++++++++++++++++++++++++
Eigenvalue #     Random Eigenvalue     Standard Dev
+++++++++++++++++++++++++++++++++++++++++++++++++++++++++
      1               1.5169               .0385
      2               1.4512               .0280
      3               1.4042               .0248
      4               1.3590               .0212
      5               1.3227               .0217
      6               1.2861               .0184
      7               1.2558               .0169
      8               1.2291               .0146
      9               1.2016               .0149
     10               1.1733               .0144
     11               1.1473               .0136
     12               1.1215               .0138
     13               1.0946               .0146
     14               1.0707               .0135
     15               1.0496               .0122
     16               1.0263               .0131
     17               1.0013               .0139
     18               0.9793               .0119
     19               0.9591               .0131
     20               0.9365               .0138
     21               0.9143               .0113
     22               0.8923               .0123
     23               0.8693               .0130
     24               0.8491               .0150
     25               0.8291               .0148
     26               0.8065               .0130
     27               0.7869               .0131
     28               0.7671               .0120
     29               0.7427               .0138
     30               0.7214               .0128
     31               0.6987               .0123
     32               0.6752               .0156
     33               0.6505               .0154
     34               0.6222               .0162
     35               0.5888               .0199
+++++++++++++++++++++++++++++++++++++++++++++++++++++++++
11/03/2017   17:50:56

Monte Carlo PCA for Parallel Analysis
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```

APPENDIX Q: Component matrix table (T0)

Component Matrix[a]

	Component										
	1	2	3	4	5	6	7	8	9	10	11
Tomatoes	.680										
Lettuce	.656										
Carrots	.595										
Peppers	.576		.302						.397		
Bananas, green bananas	.539		-.475								
Mushrooms	.525								-.336		
Citrus fruits	.508		-.356			.308	.359				
Onions (all varieties), leeks, celery	.507										
Strawberries, blackberries, cherries, etc	.502		-.365								
Eggs	.492								-.391		-.324
Apples	.489		-.348								
Herbal tea, fruit tea	.431								.425		
Yoghurt	.427					-.396					
Biscuits, chocolate or chocolate covered		.683					-.370				
Milk chocolate		.676									
Potato crisps		.622									
Biscuits, plain sweet		.620	-.355				-.404				
White bread and rolls		.569									
Butter or margarine		.557		.406							
Cakes		.556									
Potato		.428							-.351		
Crackers		.386				-.358					
Hard cheese		.384	.309			-.353					
Cooking oil (all varieties)		.406	.467								
Milk (cow's milk) (all varieties) **added to drinks**				.684							
Coffee (all varieties)				.603				-.308		-.325	
Beef			.352		.580						
Ham, bacon					.421						
Milk (cow's milk) (all varieties) **added to food**		.385			.399	-.305					
Chicken, turkey or duck			.400	-.357	.396	.406					
Black tea							-.303	.612		.356	
Milk (cow's milk) **as a drink**					.337		.330	-.401		.381	
Sugar (all varieties)				.316				-.365		.331	
Wholegrain bread and rolls										-.419	-.469
Cordials (including syrups, powders)						-.322					.356

Extraction Method: Principal Component Analysis.
a. 11 components extracted.

APPENDIX R: Rotated component matrix table (T0)

Rotated Component Matrix[a]

	Component				
	1	2	3	4	5
Milk chocolate	.696				
Biscuits, chocolate or chocolate covered	.679				
Potato crisps	.654				
Biscuits, plain sweet	.621		.324		
Cakes	.603				
White bread and rolls	.599				
Butter or margarine	.533				.436
Potato	.455	.342			
Hard cheese	.419				
Crackers	.317		.312		
Onions (all varieties), leeks, celery		.738			
Peppers (all varieties)		.652			
Onions (all varieties), leeks, celery		.605			
Lettuce		.599			
Mushrooms		.540			
Carrots		.533			
Cooking oil (all varieties)	.364	.411			
Herbal tea, fruit tea		.318	.301		
Black tea					
Bananas, green bananas			.667		
Strawberries, blackberries, cherries, blueberries, boysenberries, loganberries, cranberries, gooseberries, raspberries			.611		
Citrus fruits			.584		
Apples			.561		
Yoghurt			.551		
Beef				.675	
Chicken, turkey or duck				.640	
Ham, bacon				.551	
Eggs		.339	.321	.341	
Cordials (including syrups, powders)				.322	
Wholegrain bread and rolls					
Milk (cow's milk) (all varieties) **added to drinks**					.736
Coffee (all varieties)					.602
Milk (cow's milk) (all varieties) **added to food**			.382		.464
Sugar (all varieties)					.429
Milk (cow's milk) **as a drink**					

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.
a. Rotation converged in 7 iterations.

APPENDIX S: Total variance explained table (T1)

Total Variance Explained

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings			Rotation Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	4.760	13.600	13.600	4.760	13.600	13.600	2.429	6.941	6.941
2	1.873	5.352	18.952	1.873	5.352	18.952	1.924	5.498	12.439
3	1.710	4.886	23.838	1.710	4.886	23.838	1.910	5.457	17.896
4	1.622	4.635	28.472	1.622	4.635	28.472	1.705	4.870	22.767
5	1.553	4.436	32.908	1.553	4.436	32.908	1.666	4.759	27.525
6	1.403	4.009	36.917	1.403	4.009	36.917	1.562	4.462	31.987
7	1.297	3.707	40.624	1.297	3.707	40.624	1.548	4.422	36.409
8	1.256	3.589	44.214	1.256	3.589	44.214	1.516	4.332	40.741
9	1.195	3.413	47.627	1.195	3.413	47.627	1.457	4.162	44.904
10	1.120	3.201	50.828	1.120	3.201	50.828	1.402	4.005	48.909
11	1.101	3.147	53.975	1.101	3.147	53.975	1.257	3.592	52.500
12	1.052	3.006	56.980	1.052	3.006	56.980	1.247	3.563	56.063
13	1.022	2.919	59.900	1.022	2.919	59.900	1.196	3.418	59.481
14	1.009	2.882	62.782	1.009	2.882	62.782	1.155	3.301	62.782
15	.971	2.775	65.558						
16	.966	2.760	68.317						
17	.879	2.511	70.828						
18	.827	2.364	73.192						
19	.808	2.308	75.501						
20	.759	2.168	77.669						
21	.729	2.083	79.751						
22	.667	1.905	81.656						
23	.655	1.872	83.528						
24	.611	1.744	85.272						
25	.587	1.676	86.949						
26	.578	1.650	88.599						
27	.567	1.620	90.220						
28	.491	1.404	91.623						
29	.488	1.393	93.017						
30	.476	1.359	94.375						
31	.440	1.258	95.634						
32	.420	1.201	96.835						
33	.396	1.131	97.966						
34	.360	1.028	98.993						
35	.352	1.007	100.000						

Extraction Method: Principal Component Analysis.

APPENDIX T: Result of Monte Carlo PCA for parallel analysis (T1)

```
06/12/2017   18:24:39
Number of variables:     35
Number of subjects:     543
Number of replications: 100

++++++++++++++++++++++++++++++++++++++++++++++++++++++++
Eigenvalue #     Random Eigenvalue     Standard Dev
++++++++++++++++++++++++++++++++++++++++++++++++++++++++
      1               1.5058               .0338
      2               1.4444               .0259
      3               1.3996               .0238
      4               1.3580               .0210
      5               1.3241               .0198
      6               1.2909               .0182
      7               1.2602               .0185
      8               1.2297               .0167
      9               1.2026               .0169
     10               1.1730               .0150
     11               1.1475               .0137
     12               1.1209               .0145
     13               1.0960               .0125
     14               1.0731               .0125
     15               1.0481               .0119
     16               1.0260               .0117
     17               1.0021               .0131
     18               0.9803               .0123
     19               0.9570               .0120
     20               0.9356               .0127
     21               0.9148               .0111
     22               0.8937               .0127
     23               0.8727               .0139
     24               0.8520               .0118
     25               0.8295               .0126
     26               0.8076               .0133
     27               0.7884               .0120
     28               0.7666               .0130
     29               0.7450               .0132
     30               0.7224               .0146
     31               0.6985               .0132
     32               0.6737               .0144
     33               0.6497               .0146
     34               0.6223               .0162
     35               0.5882               .0205
++++++++++++++++++++++++++++++++++++++++++++++++++++++++
06/12/2017   18:24:48

Monte Carlo PCA for Parallel Analysis
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```

APPENDIX U: Component matrix table (T1)

Component Matrix[a]

	Component													
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Onions (all varieties), leeks, celery	.590					-.324								
Tomatoes	.578					-.385								
Lettuce	.558	-.303												
Courgette, zucchini, cucumber, gerkins or marrow (all varieties)	.553								-.301					
Corn (all varieties)	.537								-.410				-.318	
Strawberries, blackberries, cherries, blueberries, etc.	.521		-.317											
Green beans, broad beans, runner beans, asparagus	.469		.338			.437								
Citrus fruits	.467		-.346								-.306			
Broccoli	.437		.321				.400	-.314						
Mushrooms	.435					-.327								
Beetroot, radishes (all varieties)	.422	-.359												
Peas, green	.417				-.349	.359								
Peppers (all varieties)	.408					-.306								
Apples	.404										-.332			
Yoghurt	.402													
Eggs	.388											-.378		
Pears	.378							-.373						

Bananas, green bananas	.371						-.301				-.354			
Potato	.363	.577												
Pasta, noodles (white)		.425												
Grapes	.381	.384	-.351											
Chicken, turkey or duck			.519		.493									
Porridge, rolled oats, oat bran, oat meal			-.368					.322						
Milk (cow's milk) (all varieties) **added to drinks**				.713										
Black tea				.497			.327					-.472		
Coffee (all varieties)				.395			-.367	-.346						
Milk (cow's milk) (all varieties) **added to food**				.391	-.315			.320						.346
Ham, bacon				.334	.322									
Beans in sauce	.362	.406			-.408									
Beef (eg. roast, steak, chops, silverside, casseroles, stir fry, curry, mince dishes)				.351	.354									
Spinach, silver beet, swiss chard	.303						-.491						.346	
Stone fruit	.382						.437							
Herbal tea, fruit tea									.456					
Cordials (including syrups, powders)									-.446				.357	
Wholegrain bread and rolls										.770				

Extraction Method: Principal Component Analysis.
a. 14 components extracted.

APPENDIX V: Ethical approval letter from the University of Nottingham for study 2

The University of Nottingham

Direct line/e-mail
+44 (0) 115 8232561
FMHS-ReschEthics@nottingham.ac.uk

Faculty of Medicine and Health Sciences

Research Ethics Committee
C/o Faculty PVC Office
School of Medicine Education Centre
B Floor, Medical School
Queen's Medical Centre Campus
Nottingham University Hospitals
Nottingham
NG7 2UH

7th November 2016

Dr Fiona McCullough
Associate Professor
Nutritional Sciences
School of Biosciences
Sutton Bonington Campus
University of Nottingham
Loughborough
Leics, LE12 5RD

Dear Dr McCullough

Ethics Reference No: OVS18102016 **– please always quote**	
Study Title: Assessing the relationship between iron status, dietary iron intake and body mass index in female adults whose weight is greater than the healthy weight range.	
Chief Investigator/Supervisor:	
Lead Investigators/student: Husna Kaya Kacar, PhD Student, Nutritional Sciences.	
Other Key Investigators: Amanda Avery, Assistant Professor, Nutritional Sciences.	
Type of Study: PhD Project , Overseas - Turkey	
Proposed Start Date: 01.12.16	**Proposed End Date:** 30/06/2017 6mths
No of Subjects: 100	**Age:** 18-49 years
School: Biosciences	

Thank you for submitting the above application which was considered by the Committee at its meeting on 18th October 2016 and the following documents were received:

- FMHS Research Ethics Application dated 17.10.16
- Study Protocol
- Appendix 1: Research Ethics approval process in Turkey.
- Healthy Volunteer's Information Sheet
- Consent Form
- Weight and Iron Status – Are the Two Linked Questionnaire
- Participant Record Card.
- Turkish Public Hospital Association Research Ethics Application Form.

These have been reviewed and are satisfactory and the study has been given a favourable opinion.

A Favourable opinion is given on the understanding that all appropriate ethical and regulatory permissions are respected and followed in accordance with all local laws of the country in which the study is being conducted and those required by the host organisation/s involved.

A favourable opinion is given on the understanding that the conditions set out below are followed:

1. Please submit copies of letters of permission/approval from the Turkish Public Hospital Association, the Hospital Administrators at the Giresun State Hospital, Giresun and Bafra State Hospital, Samsun, Turkey when available for our records.

The University of Nottingham

2. You must follow the protocol agreed and inform the Committee of any changes using a notification of amendment form (please request a form).
3. You must notify the Chair of any serious or unexpected event.
4. This study is approved for the period of active recruitment requested. The Committee also provides a further 5 year approval for any necessary work to be performed on the study which may arise in the process of publication and peer review.
5. An End of Project Progress Report is completed and returned when the study has finished (Please request a form).

Yours sincerely

pp Lamb

Professor Ravi Mahajan
Chair, Faculty of Medicine & Health Sciences Research Ethics Committee

APPENDIX W: Ethical approval letter from the Ministry of Health (Turkey) for study 2

SAMSUN İLİ KAMU HASTANELERİ BİRLİĞİ GENEL SEKRETERLİĞİ - SAMSUN KHBGS İDARİ HİZMETLER BAŞKANLIĞI
23/12/2016 11:22 - 54103609 - 604.02 - E.23329
00035914444

T.C.
SAĞLIK BAKANLIĞI
Türkiye Kamu Hastaneleri Kurumu
Samsun İli Kamu Hastaneleri Birliği Genel Sekreterliği

2017 SAMSUN DEAFLYMPICS

Sayı : 54103609-604.02
Konu : Araştırma İzni (Hüsna KAYA KAÇAR)

DAĞITIM YERLERİNE

İlgi : 14/12/2016 tarihli ve 5794 sayılı dilekçe;

İlgi tarih ve sayılı dilekçeye istinaden; Nottingham Üniversitesi Beslenme Bilimleri doktora öğrencisi Hüsna KAYA KAÇAR'ın "Vücut Ağırlığı ve Demir Düzeyi" konulu araştırmalarını hastanenizdeki 19-49 yaş arası, diyetisyene ilk defa başvuran bayanlara yapmaları uygun görülmüş olup, araştırmanın yapılabilmesi için ilgili kişi ile "Araştırma İzinleri İşbirliği Protokolü" imzalanarak ek'te sunulmuştur.

Bilgilerinizi ve gereğini rica ederim.

Dr. Yılmaz DÜNDAR
Genel Sekreter a.
İdari Hizmetler Başkanı

EK:
1. Araştırma İzinleri İşbirliği Protokolü
2. Araştırma Çalışmaları Başvuru Formu
3. Anket Formu (Kitapçık olduğu için zarfla gönderilecektir)

DAĞITIM:
Gereği :
1-Bafra Devlet Hastanesi
2-Hüsna KAYA KAÇAR
KaraSamsun Mahallesi
Yıldıray Çınar Bulvarı Seymen Apt.
No:65/9 İlkadım/SAMSUN

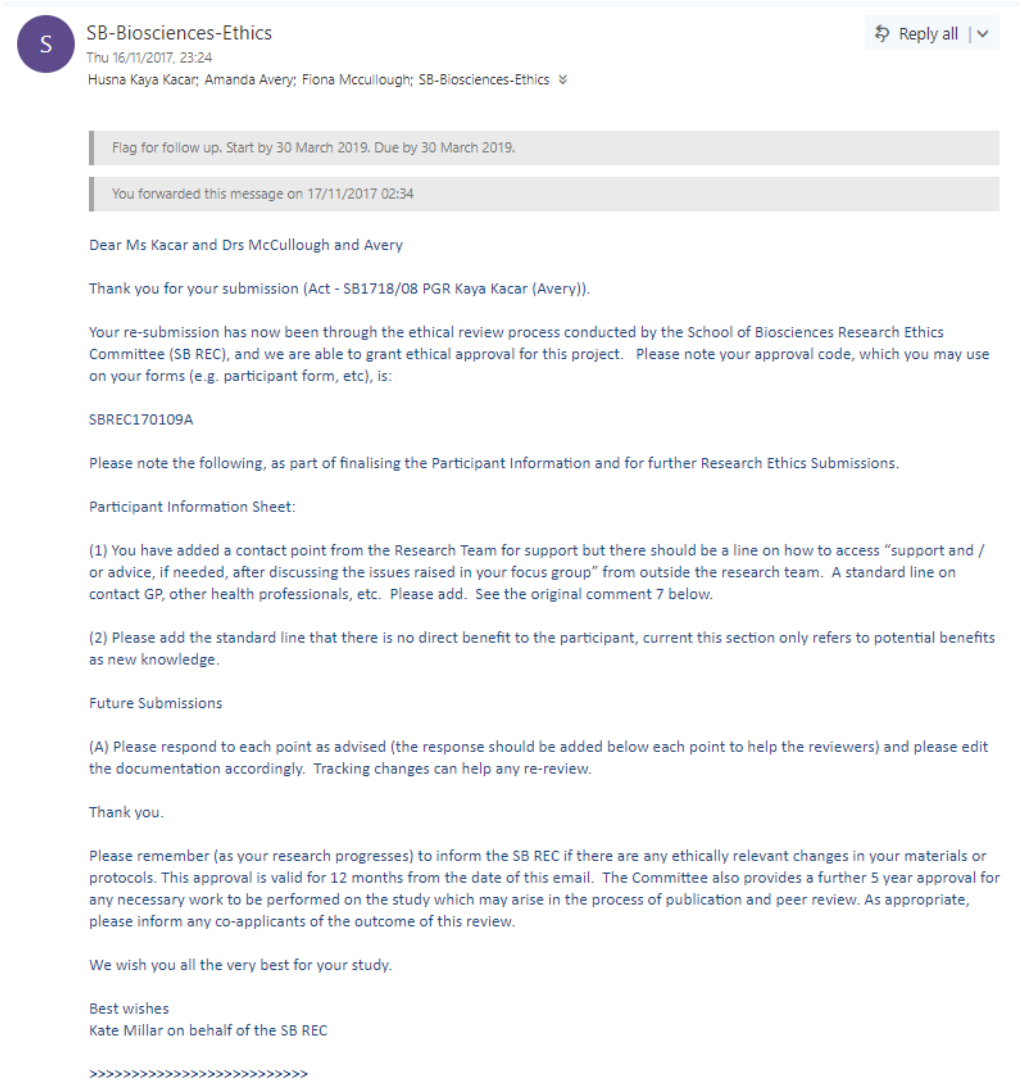
Samsun Kamu Hastaneleri Birliği Genel Sekreterliği/ Merkezi
Bulvarı No:232 İlkadım/SAMSUN
311 2500 (1428)
Faks No:0(362) 311 25 28
e-Posta:kursat.yurdakos@saglik.gov.tr İnt.Adresi: -

Araştırma ve Geliştirme (Ar-Ge)
Adalet Mahallesi 100.Yıl
(0362)

Bilgi için:KÜRŞAT YURDAKOŞ
Unvan:UZMAN
Telefon No:0362 311 25 00/1428

Evrakın elektronik imzalı suretine http://e-belge.saglik.gov.tr adresinden dd6aa74c-08b5-41a0-ba09-6a773ae6a362 kodu ile erişebilirsiniz.
Bu belge 5070 sayılı elektronik imza kanuna göre güvenli elektronik imza ile imzalanmıştır.

APPENDIX X: Ethical approval e-mail (qualitative study)



S

SB-Biosciences-Ethics

Thu 16/11/2017, 23:24

Husna Kaya Kacar; Amanda Avery; Fiona Mccullough; SB-Biosciences-Ethics

Reply all

Flag for follow up. Start by 30 March 2019. Due by 30 March 2019.

You forwarded this message on 17/11/2017 02:34

Dear Ms Kacar and Drs McCullough and Avery

Thank you for your submission (Act - SB1718/08 PGR Kaya Kacar (Avery)).

Your re-submission has now been through the ethical review process conducted by the School of Biosciences Research Ethics Committee (SB REC), and we are able to grant ethical approval for this project. Please note your approval code, which you may use on your forms (e.g. participant form, etc), is:

SBREC170109A

Please note the following, as part of finalising the Participant Information and for further Research Ethics Submissions.

Participant Information Sheet:

(1) You have added a contact point from the Research Team for support but there should be a line on how to access "support and / or advice, if needed, after discussing the issues raised in your focus group" from outside the research team. A standard line on contact GP, other health professionals, etc. Please add. See the original comment 7 below.

(2) Please add the standard line that there is no direct benefit to the participant, current this section only refers to potential benefits as new knowledge.

Future Submissions

(A) Please respond to each point as advised (the response should be added below each point to help the reviewers) and please edit the documentation accordingly. Tracking changes can help any re-review.

Thank you.

Please remember (as your research progresses) to inform the SB REC if there are any ethically relevant changes in your materials or protocols. This approval is valid for 12 months from the date of this email. The Committee also provides a further 5 year approval for any necessary work to be performed on the study which may arise in the process of publication and peer review. As appropriate, please inform any co-applicants of the outcome of this review.

We wish you all the very best for your study.

Best wishes
Kate Millar on behalf of the SB REC

>>>>>>>>>>>>>>>>>>>>>>>>>