Timeline over which compensatory mechanisms are activated during weight loss with a very-low-calorie diet.

Jeanette Jørgensen

Master Thesis in Clinical Health Science

May 2014

Norwegian University of Science and Technology

Faculty of Medicine

Institute of Public Health and General Practice

NTNU

Norwegian University of
Science and Technology

Abstract

Background: Diet-induced weight loss (WL) activates several compensatory mechanisms, both at the level of energy intake (EI) and energy expenditure (EE), which increases the risk of relapse. Unfortunately little is known regarding when these mechanisms are activated and how they respond with progressive WL.

Purpose: To assess the timeline over which compensatory mechanisms are activated during WL with a very-low-calorie diet (VLCD).

Material & methods: Twelve healthy obese men and women underwent an 8-week VLCD. Body weight and composition, resting energy expenditure (REE), exercise efficiency (at 10, 25 and 50 W) and subjective feelings of appetite (in fasting and for 2.5 h after a meal) were measured at baseline, day 3, after 1 week, when each individual participant lost 5 % and 10 % of his/her baseline weight and at the end of the intervention.

Results: Significant WL was seen at day 3 and got stronger with progressive WL. REE was significantly reduced at 5 % WL and remain stable until end of study. A significant increase in exercise efficiency at 10 W was only seen at end of VLCD (average WL of 18.4±2.3 kg). No significant changes in subjective feelings of appetite were observed, but there was a tendency (P=0.058) for an increase in subjective feelings of hunger in fasting at day 3.

Conclusion: Losing weight with a VLCD (ketogenic diet) leads to a significant reduction in REE from 5 % WL with no changes afterwards, while an increase in exercise efficiency at 10 W is only seen at the end of the intervention. Subjective feelings of appetite do not seem to change significantly over time, even though a slight increase in fasting hunger may be seen at day 3.

Relevance

The assessment of the timeline over which compensatory mechanisms, both at the level of EI and EE, are activated with WL is extremely important to health professionals working within obesity management. This knowledge will provide health professional with the understanding of when compensatory mechanisms start to be activated and also, potentially, when a plateau is reached. This information is very valuable since it can be used to try to help obese people to lose weight and maintain it in the long term.

Acknowledgements

I would like to thank my supervisor, Associate Professor Catia Martins. I am forever grateful for her help throughout this last year. Her guidance, quick feedback and wisdom have been indispensable during the work of this thesis.

A special thanks to Hege Tevik Bjøru, Linn Bøhler, Sìlvia Coutinho, Sissel Salater, Linn-Christin Torgersen and Emilie Føyen With for help with implementation of the study. Without their help this study would never have been conducted. Thank you!

Last, but not least, I would like to thank all study participants for their time and patience, Turid Follestad for her statistical advice, and friends and family for support. I am forever grateful!

Jeanette Jørgensen

Table of Contents

1. Background

Over 1.4 billion people worldwide were overweight, and almost 500 million of those were obese in 2008 (1). The prevalence of obesity is increasing, and Norway is no exception (1-4). A health survey done over 22 years in Norway revealed that obesity (BMI≥30 kg/m²) prevalence has increased from 7.7 % in 1984-86 to 22.1 % in 2006-08 in men and from 13.3 % to 23.1 % in women (4). Obesity is thought to contribute for up to 8 % of health budgets, but it also costs employers, tax payers and society. For the individual, obesity can result in struggle with illness, disability and early death (5). Fortunately, research over the latest years has shown that a WL between 5-10 % of baseline weight, if sustained, can have huge health benefits by preventing or improving several obesity-related risk factors and comorbidities (such as cardiovascular diseases, diabetes, musculoskeletal disorders and some cancers) (1, 6). The first hand treatment of obesity should be lifestyle interventions that include diet, exercise and behavior modification (7). However, most people will experience significant weight regain in the long-term (7-9). It is therefore important to understand the mechanisms behind weight regain, so relapse can be minimized or prevented.

Compensatory mechanisms activated during WL with VLCD

People seem to struggle with relapse after WL due to metabolic, behavioral, neuroendocrine and autonomic responses that try to revert body energy stores to their original levels (10). These mechanisms are activated both at the level of EE and EI (11). Diet-induced WL is associated with a reduction in total energy expenditure (TEE), REE and non-resting energy expenditure (NREE), as well as a decrease in fat oxidation due to changes in substrate metabolism, and an increase in exercise efficiency (10-15).

TEE consist of REE (the energy needed to fuel minimal daily functions of cells and organs; approximately 60-70 % of TEE) and NREE (energy expended above resting level; approximately 30-40 % of TEE) (10). A study, where a 10 % WL was achieved with energy restriction, showed a mean reduction in TEE of 8±5 kcal/kg/day (16). The same study indicated a decrease of 3-4 kcal/kg of fat-free mass (FFM) in REE and NREE (16).

Diet-induced WL is usually followed by a reduction in REE, as a result of loss in FFM (17). However, some studies have shown much larger reduction in REE than what could be

predicted from loss of FFM and fat mass (FM), a process known as adaptive thermogenesis (12, 18).

WL of 10 % or more, in previously obese individuals, has been shown to result in a significant reduction in NREE, more than expected for the given reduction in FFM and FM (16). The impact of WL on physical activity (PA) levels remains controversial, with some showing a reduction (19, 20) and other and increase (21). Despite this, a reduction in activity energy expenditure (AEE) is the usual outcome (19, 21), even with increased PA levels (21). This is likely to be due to an increase in exercise efficiency with WL, meaning that exercise-induced EE for a given volume of exercise is reduced. Several studies have found that diet-induced WL in obese people is associated with increased exercise efficiency, especially at lower intensity levels (14, 22-24).

Several studies have also shown that diet-induced WL is associated with a change in substrate oxidation towards a reduction in fat oxidation (12, 25, 26).

Moreover, compensatory mechanisms are also activated at the level of EI. Diet-induced WL seems to increase feelings of hunger and decrease feelings of fullness, even though EE declines (10, 11, 27-30). Different appetite hormones are involved in appetite control, and identified appetite modulators includes leptin, ghrelin, cholecystokinin (CCK), peptide YY (PPY), insulin, pancreatic polypeptide (PP), and glucagon-like peptide 1 (GLP-1). Leptin has a role in reducing food intake and increasing EE, while ghrelin stimulates hunger, and CCK, PPY and GLP-1 seem to inhibit food intake in the postprandial period (satiety hormones). Diet-induced WL is usually associated with reduced circulating levels of leptin (26, 28, 31), increased fasting plasma levels of ghrelin (28, 31-33) and reduced plasma levels of satiety hormones (PPY, CCK and GLP-1), both in fasting and postprandial period (28, 34-36). The changes in appetite hormones after WL make people feel hungrier and less satiated after they have lost weight. Interestingly, the changes in appetite-related hormones observed with WL seem to persist in the long-term (1 year), even with partial weight regain (28). The compensatory mechanisms described before are likely to increase the risk of weight regain, making relapse almost inevitable.

Timeline of compensatory mechanisms

Few studies have managed to isolate the effects of energy restriction and weight reduction in terms of activation of compensatory mechanisms (6), and none has looked at the timeline over which these mechanisms are activated. One study in overweight individuals showed a significant increase in ghrelin concentrations, reduction in leptin concentrations, and reduction in subjective feeling of fullness after only 2 days of energy restriction, before any significant WL had been achieved (37). A decline in leptin and increase in hunger feelings has also been reported after 4 days of a 65 % energy restricted diet, despite only a small, but significant WL (2.4 kg) in overweight participants (38). Conversely, another study in overweight men, who followed 4 days of a 800 kcal/day diet reported no change in fasting or postprandial total ghrelin plasma levels, despite a significant decrease in leptin and a significant WL of 1.3 kg (39).

Moreover, a study where participants followed a VLCD for eight weeks, showed a 17 % decrease in REE after the first 2 weeks (average 5.8 % WL), but there was a significant increase in REE afterwards when energy restriction was 1200 kcal/d, despite continued WL (40). Another study with 8 weeks of energy restriction found that REE was significantly reduced after 2 weeks with an average WL of 1.5 kg in men and 0.9 kg in women, and REE was reduced further at week 8 in both women and men (WL of 5.1 kg and 8.7 kg, respectively) (41). Therefore, it is important to assess the timeline over which compensatory mechanisms, both at the level of EI and EE, are activated when obese people lose weight. Do these compensatory mechanisms get stronger over time with progressive WL or do they reach a plateau after a specific WL? And are they activated before any significant WL is achieved or would a minimal WL be needed in order to initiate these mechanisms?

2. Aim

The overall aim for this thesis was to assess the timeline over which compensatory mechanisms, at level of both EI and EE, are activated during WL with an 8-week VLCD. Secondary aims were to see if those compensatory mechanisms got stronger with progressive WL or reached a plateau after certain WL, and if they were activated, even before any significant WL was achieved. Moreover, it was also aimed to assess if there were gender differences in the activation of compensatory mechanisms during WL.

The main hypothesis for this thesis was that the strength of the compensatory mechanisms activated during WL reach a plateau after a certain reduction in body weight, and that men have stronger and earlier activation of compensatory mechanisms than women.

3. Material and Methods

3.1 Study design

This is a longitudinal study with repeated measurements, where participants served as their own controls.

3.2 Participants

Fourteen healthy obese volunteers were recruited to this study through advertisement posted on the intranet of NTNU, St. Olav's hospital and local newspaper. Inclusion criteria were age between 18-50 years old, a BMI between 30-47 kg/m², weight stable in the last 3 months (not more than 2 kg variation), not currently dieting to lose weight and with an inactive lifestyle. Due to the potential impact of phase of menstrual cycle of the outcome variables (42, 43), it was decided to include only men, post-menopausal women and women taking hormonal contraceptives. Exclusion criteria included pregnancy, breastfeeding, drug- or alcohol abuse within the last two years, medication known to affect appetite or induce WL, and participation in another obesity treatment program. Moreover, those with a history of psychological disorders, eating disorders, diabetes 1 or 2, gastrointestinal (especially colelithiasis), kidney, liver, lung, cardiovascular disease and malignancies were not approved to take part in this study. All participants gave a written informed consent, the data was anonymous and treated confidential, and the study was approved by the local ethical committee (Ref., 2012/1901).

3.3 Procedure

Participants underwent a commercial VLCD (Allevo, Cederroth, Sweden) (550 kcal/day for women and 660 kcal/day for men) for 8 weeks. The aim was that participants lost on average 14 % of their baseline weight, an amount that has been reported in comparable interventions and has been found to be safe (28, 44, 45). Expected WL was calculated using a body weight simulator (45). Mean age of 34 years and mean BMI of 38.5 kg/m² were used in the calculations, but also a light activity level (PA level of 1.6), and kcal/day was set to 550 for women and 660 for men. The body weight simulator gave a mean expected WL on 13.6 % in women and 14.8 % in men after 8 weeks on VLCD. The VLCD products provide 110-120

kcal/pack depending on which products the participants selected. The participants could choose between shakes (110 kcal/portion) (banana & raspberry, chocolate or strawberry & blueberry flavor) and soups (110-120 kcal/portion) (broccoli & basil soup with couscous or potato & leek soup). Each product was designed to provide high levels of proteins supplemented with vitamins, minerals, and fatty acids. Women had to drink/eat five products per day, and men six, and the participants were allowed to have calorie-free drinks, low-starch vegetables (e.g. broccoli, cauliflower, tomatoes, cucumber or lettuce) (maximum 100 g/day), sugar-free gum and soda, maximum 4 sugar-free pastilles and were recommended to drink minimum 2.5 liters of non-caloric liquids. After completion of the commercial VLCD, a dietician recommended an individual diet matched for energy needs to every participant with the aim of WL maintenance.

3.4 Compliance

To check for compliance with the intervention, participants were requested to keep daily food records and had a weekly follow up with the researcher for weighing, measurement of ketone bodies in urine (using Ketostix (Bayer, Basel - Switzerland)) and discussion of food records. The participants received VLCD-products on each weekly visit and side-effects, if present, were also registered. Negative ketone bodies more than once during the 8 weeks resulted in exclusion from the study.

During the study, participants were asked not to change their PA levels. As a measure of compliance for maintenance of PA levels throughout the study, participants were asked to use arm bands (BodyMedia®, SenseWear, Pittsburgh - USA) on the back of the upper triceps (right-handed wore the armband on their left arm and vice versa), for one week at baseline, week 1, 4 and 8 of the intervention. The SenseWear Armband measures PA by a biaxial accelerometer and a combination of heat sensors, and provides information on average metabolic equivalent (MET), time spent on PA at different intensities, steps per day, TEE and AEE. Only a few participants had 7 days of valid data (5 weekdays and 2 weekend days), therefore four days (3 weekdays and 1 weekend) was used as the minimum accepted number of days. Each day had to have at least 1368 min of data (95 % of 24 h) to be valid. The following cut points were used to look at PA of different intensities in this obese patient population: sedentary (<1.5 MET), light (1.5-3 MET), moderate (3-6 MET) and vigorous (>6 MET) activity (46-48).

3.5 Measurements

The presence and strength of compensatory mechanisms were assessed at baseline, day 3, after 1 week, and when each participant lost 5 % and 10 % of their baseline body weight and at the end of the intervention (week 9). The following measurements were performed at the 6 time points previously described: Body weight and composition, REE, substrate oxidation, exercise efficiency and subjective feelings of appetite. For a detailed description of each measurement see below:

Anthropometric measurements

Weight and height were measured using a scale (Seca, Gmbh & co, Germany) and a stadiometer (Seca, Gmbh & co, Germany) to the nearest 0.1 kg and 0.5 cm, respectively. All participants were asked to empty their bladder beforehand and were weighed in underwear. The height was measured without shoes, heels against the wall, and a straight body with the head in a normal position.

Body composition

Body composition (body fat percent (% BF), FM in kg, FFM in percent (% FFM) and FFM in kg) was measured in fasting using Air Displacement Plethysmography (BodPod, COSMED, Italy). The BodPod measures body composition from a whole-body densitometry by an inverse relationship between pressure and volume inside the machine, with appropriate corrections for the air in lungs and gut. The participants wore underwear and a bathing cap (to minimize volume of air around the hair) inside the machine.

REE

REE and fasting substrate oxidation (respiratory quotient – RQ) were measured using indirect calorimetry (Vmax Encore 29 N, CareFusion, Germany). The participants were in fasting (at least 10 hours fasting), and were asked not to smoke, drink anything except water from the night before and not to exercise before the test. The participants were asked to sit on a chair upon arrival for at least 10 minutes and after that to lie down on a bench. A canopy was then placed over their head and oxygen uptake (VO_2), carbon dioxide (VCO_2) consumption measured continuously for at least 15 minutes (or as long as needed to obtain at least 10 minutes of stable data), after excluding the first five minutes (49).

Exercise efficiency

Exercise efficiency was measured using a graded cycle ergometry (Monark, Eromedic 839E, GIH, Sweden). First the participants had a 5-min period of a comfortable warm-up, followed by pedaling at 60 rpm against graded resistance to generate 10, 25 and 50 W of power in sequential 4-min intervals. VO_2 consumption, VCO_2 production and RQ were measured continuously using a metabolic cart (Vmax Encore 29N, CareFusion, Germany). Exercise efficiency was expressed as gross mechanical efficiency, which is defined as power output /power input ((kcal/min, 1W=0,01433kcal/min)/(VO_2-average VO_2 during steady state period (lasts 2 minutes of each stage) times the oxygen equivalent))(14).

Subjective feelings of appetite

Subjective feelings of appetite (hunger, fullness, desire to eat and prospective food consumption (PFC)) were assessed by visual analogue scales (VAS) (50) in fasting, immediately after a standardized breakfast and every 30 minutes up to 150 minutes. Breakfast consisted of 75 g of bread, 150 g orange juice, 250 g milk with 1.5 % fat, 5 g TINE dairy butter, 35 g TINE Gräddost cheese and 40 g NORA strawberry jam (total of 596.6 kcal, 15.6 % protein, 35 % fat and 48.1 % carbohydrate). Milk could be replaced with 250 g of cherry yoghurt drink, but then the participant only got 100 g of orange juice and 20 g of jam (total of 604.1 kcal, 15 % protein, 33.9 % fat and 50 % carbohydrate). The participant had to answer 4 questions every time point by putting a cross on a 10 centimeter long line. The questions were:

1. How hungry do you feel? (not hungry at all - never been hungrier)
2. How full do you feel (not full at all - very full)
3. How much food do you think you can eat? (nothing - a lot)
4. How much food do you want to eat? (nothing - a lot)

3.6 Sample size calculation

The sample size estimation was based on changes in REE over time (day 3: -10; week 1: - 50, 5 % WL: -100; 10 % WL: -180 and end: -250 Kcal/day) (18), assuming an SD of 229 kcal/day and a low correlation (r=0.3) in REE between time points. Twelve participants would be necessary, given a power of 80 %, a significance level of 5 %, and a predicted drop-out rate of around 10 %.

3.7 Statistics

Statistical analysis was performed using IBM SPSS statistics 21 (SPSS Inc., Chicago, IL) and statistical significance assumed at $P<0.05$, except when required otherwise. Normal distribution was checked by the Shapiro Wilk Test and visual inspection of Q-Q plots. Differences between men and women were checked by independent-sample T-test for normal distributed data and Mann-Whitney U Test for non-parametric data. Descriptive statistics (means and standard deviations (SD)) were computed for all variables, and a repeated measure ANOVA, with Bonferri adjustment for multiple comparisons, was used to look at changes in the variables of interest over time in normal distributed data. Gender differences were checked by running a mixed ANOVA, with Bonferri adjustment for multiple comparisons, with time as a within-subjects variable and gender as a between-subjects variable. Not-normal distributed data was analyzed using Friedman's ANOVA Test. Area under the curve (AUC) was calculated using the trapezoid rule.

4. Results

4.1 Study population

Of the fourteen participants who started, two subject dropped out, one due to personal reasons and one due to side effects (vomiting, dizziness and fatigue) from the VLCD. The final study population was composed of 12 participants (7 women and 5 men). Baseline characteristics of all 12 participants are presented in **Table 1**.

Table 1. **Baseline characteristics of participants.**

	All (n=12)	Men (n=5)	Women (n=7)	P-value *
Age	45.4 ± 7.2	43.6 ± 4.6	46.7 ± 8.7	NS
Height (m)	1.71 ± 0.1	1.77 ± 0.1	1.66 ± 0.1	0.056
Weight (kg)	110.6 ± 18.0	121.9 ± 14.0	104.7 ± 19.0	NS
BMI (kg/m²)	38.3 ± 4.4	39.1 ± 3.4	37.8 ± 5.2	NS

Data presented as mean±SD. * Gender differences. NS=Non significance.

There were no significant differences in age, weight, and BMI between genders, but there were a borderline significance in height between genders ($P<0.056$). Regarding women; two were post-menopausal and five used hormonal contraceptives.

4.2 Compliance with the intervention

Three participants had negative urine ketone bodies on one occasion during the 8 weeks of VLCD.

PA Armband

Five participants had valid armband data for all 4 time points. There were no statistically significantly differences in the mean score in MET, sedentary time (min), light activity (min), moderate activity (min), total PA time (min), steps per day or AEE (kcal) over time.

4.3 Side effects

All participants experienced bad breath (100 %) and 42 % experienced dizziness, 42 % constipation, 33 % fatigue, 25 % increased chills, 25 % dry skin and 8 % poorer sleep, diarrhea, or vomiting.

4.4 Body weight and body composition

Body weight was significantly reduced in all 12 participants during 8 weeks on VLCD and between all 6 time points (**Table 1 and Figure 1**). The average WL for all participants after 8 weeks was 16.4 %. Women had an average significant WL of 15.3 % and men 17.8 %.

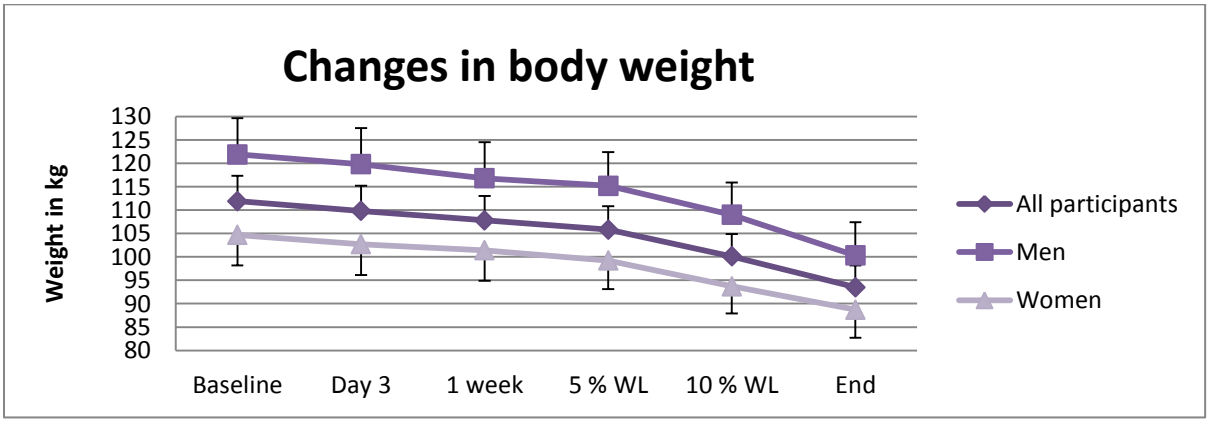

Figure 1. Changes in body weight in all participants, men and women, during 8 weeks of VLCD. Data presented as mean± SEM.

Body weight was reduced with an average of 18.4±2.3 kg during 8 weeks on VLCD and men had a higher average mean body weight than women throughout the study. WL at 5 % of body weight was achieved on average at day 14±4 (women at day 15±5 and men day 12±4)

and 10 % WL on day 32±6 (women at day 36±5 and men day 28±5). Changes in body weight and body composition over time can be seen in **Table 2**.

Table 2. Changes in body weight and body composition over time.

	Baseline	Day 3	1 Week	5 % WL	10 % WL	End	P-value
Weight,kg	111.9±18.6	109.8±18.7	107.8±18.1	105.8±17.5	100.1±16.6	93.5±16.3	<0.0001a)
Men	121.9±14.0	119.8±14.6	116.8±14.4	115.2±13.1	109.0±12.8	100.3±14.2	NSb)
Women	104.7±19.0	102.7±18.9	101.4±18.7	99.2±17.9	93.7±16.9	88.7±16.9	
FM, kg	50.2±11.3	49.0±11.2	47.7±11.1	46.2±10.4	41.6±9.6	36.4±9.7	<0.0001a)
Men	48.4±10.4	47.5±10.1	46.0±10.5	44.9±9.4	39.6±8.6	33.9±8.6	NSb)
Women	51.4±12.6	50.1±12.6	48.9±12.1	47.2±11.7	43.0±10.7	38.2±10.7	
% FM	44.8±6.3	44.6±6.1	44.1±6.1	43.6±6.1	41.5±6.4	38.7±6.5	<0.0001a)
Men	39.3±5.0	39.4±4.6	39.0±5.0	38.6±4.8	35.9±4.8	33.4±4.9	NSb)
Women	48.7±3.7	48.3±3.9	47.8±3.8	47.2±4.1	45.4±3.9	42.5±4.7	
FFM, kg	61.7±12.2	60.8±11.9	60.1±11.3	59.7±11.4	58.6±11.5	57.1±10.5	<0.0001a)
Men	73.6±5.8	72.2±6.2	70.6±5.1	70.4±5.8	69.6±5.9	66.4±6.5	<0.0001b)
Women	53.2±7.0	52.6±6.9	52.5±7.3	52.0±7.3	50.8±9.9	50.5±7.1	
% FFM	55.2±6.3	55.4±6.1	55.9±6.1	56.4±6.1	58.5±6.4	61.3±6.5	<0.0001a)
Men	60.7±5.0	60.6±4.6	61.0±5.0	61.4±4.8	64.1±4.8	66.6±4.9	NSb)
Women	51.3±3.7	51.7±3.9	52.2±3.8	52.8±4.1	54.6±3.9	57.5±4.6	

Data presented as mean±SD. NS= Non significance. A significant main effect of time was found on body weight and body composition and a main effect on gender for FFM (kg) only. **a)** Significance for main effect of time. **b**) Significance for main effect of gender.

A significant main effect of time (P<0.0001) and a time*gender interaction (P<0.01) was found on body weight, but no main effect on gender. Further Post Hoc analysis revealed that body weight was different between all time points (P<0.001 (between 1 week and 5 % WL P<0.01) and in men, there was no significant decline in WL between 1 week and 5 % WL (P>0.05).

A significant main effect of time (P<0.0001) was found on FM in kg with a significant reduction of 13.8±1.6 kg after 8 weeks on VLCD, but no significant main effect of gender or time*gender interaction. Further Post Hoc analysis revealed that FM in kg was not significantly different between week 1 and 5 % WL (a borderline (P=0.068), but significant differences were observed among all other time points (P<0.01).

A significant main effect of time (P<0.001) and gender (P<0.01) was found on % BF, but no significant gender*time interaction. Body fat (BF) decline by an average of 6 % and was higher in women (average 9 % more) compared with men. Further Post Hoc analysis revealed that % BF was significant different between end and all time points (P<0.001), and between 10 % WL and all time points (P<0.001).

FFM reduced on average 4.6±1.7 kg after 8 weeks on VLCD. A significant main effect of time (P<0.0001) and gender (P<0.0001) and a time*gender interaction (P<0.0001) was found on FFM in kg. FFM was significantly higher in men compared with women (average difference of 18.6±13.4 kg). Further Post Hoc analysis revealed that FFM in kg was not significantly different between day 3 and 1 week and 1 week and 5 % WL, but was significant between all other time points (P<0.01) (P<0.05 between baseline and day 3 and between day 3 and 5 % WL). However, women did not have a significant declined in kg FFM until 10 % WL (P<0.001), while men had a significant decline after 1 week (P<0.01).

The percentage of FFM declined on average 6 % in all participants. A significant main effect of time (P<0.001) and gender (P<0.01) were found on % FFM, but no significant time*gender interaction. The mean differences between genders in % FFM were 9 % where men had more % FFM than women. Further Post Hoc analysis revealed that % FFM was different between 10 % WL and all time points (P<0.0001) and between end and all time points (P<0.0001).

4.5 REE and RQ

Changes in REE and RQ over time can be seen in **Table 3**.

Table 3. Changes in REE and RQ over time

	Baseline	Day 3	1 week	5 % WL	10 % WL	End	P-value
REE,kcal/day							
All	1498±208	1438±230	1449±220	1349±274	1286±194	1251±180	<0.0001a)
Men	1709±103	1629±186	1629±100	1579±121	1456±96	1359±145	<0.001b)
Women	1348±96	1302±148	1321±190	1184±228	1164±147	1174±169	
RQ							
All	0.86±0.05	0.77±0.04	0.75±0.02	0.75±0.04	0.76±0.04	0.76±0.02	<0.0001a)
Men	0.86±0.04	0.79±0.03	0.75±0.02	0.77±0.05	0.76±0.04	0.77±0.02	NSb)
Women	0.85±0.05	0.76±0.04	0.76±0.02	0.74±0.03	0.76±0.04	0.75±0.02	

Data presented as mean±SD. NS= Non significance. A significant main effect of time and gender were found for REE and for RQ a significant main effect of time. **a)** Significance for main effect of time. **b**) Significance for main effect of gender.

A significant main effect of time (P<0.0001) and gender (P<0.001) were found on REE, but no significant time*gender interaction. REE declined by an average of 247±28 kcal (P<0.001) from baseline to end of the study and men had an average higher REE (311±240 kcal per day) (P<0.001) compared with women. Further Post Hoc analysis revealed that REE was different between baseline and 5 % WL (P<0.01), 10 % WL (P<0.0001) and end (P<0.0001), between day 3 and 10 % WL (P<0.001) and end (P<0.001), and between 1 week and 5 % WL (P<0.05), 10 % WL (P<0.01) and end (P<0.01). A significant main effect of time (P<0.0001) was found on RQ, but no significant effect on gender or time*gender interaction. Further Post Hoc analysis revealed that RQ was different between baseline and the other 5 time points (P<0.01).

4.6 Exercise efficiency and RQ

Changes in Exercise efficiency and RQ over time are shown in **Table 4**.

Table 4. Changes in exercise efficiency and RQ over time.

	Baseline	Day 3	1 week	5 %WL	10 %WL	End	P-value[a])
Ex.eff*-10W	0.048±0.012	0.050±0.012	0.049±0.01	0.052±0.010	0.052±0.011	0.057±0.013	<0.05
Ex.eff*-25 W	0.097±0.021	0.103±0.021	0.101±0.015	0.105±0.016	0.105±0.014	0.108±0.019	NS
Ex.eff*-50 W	0.142±0.021	0.146±0.020	0.142±0.016	0.147±0.015	0.149±0.013	0.151±0.017	NS
RQ- 10 W	0.90±0.34	0.83±0.05	0.80±0.07	0.79±0.05	0.79±0.05	0.80±0.06	<0.001
RQ- 25 W	0.88±0.04	0.83±0.04	0.80±0.05	0.79±0.04	0.79±0.06	0.80±0.06	<0.001
RQ- 50 W	0.90±0.05	0.86±0.05	0.83±0.05	0.83±0.05	0.83±0.07	0.84±0.07	<0.05

Data presented as mean±SD. *Ex.eff=Exercise efficiency. NS=Non significance. A significant main effect of time, but no effect on gender or time*gender interaction were found for exercise efficiency and RQ. **[a])** Significance for main effect of time.

A significant main effect of time (P<0.01) was found on exercise efficiency at 10 W, but no main effect on gender or time*gender interaction. A significant increase in exercise efficiency at 10 W was observed only at end of the intervention (P<0.05). No significant main effect of time, gender or time*gender interaction were found on exercise efficiency at 25 W and 50 W.

A significant main effect of time (P<0.001) was found on RQ (10-50W), but no main effect on gender or time*gender interaction. RQ during exercise declined from baseline to end at all 3 different resistances (10-50 W). Further Post Hoc analysis revealed statistical significant differences in RQ at 10 W and 25 W between baseline and the other 5 time points (P<0.05 and P<0.001, respectively), but also between day 3 and 5 % WL at 10 W (P<0.05) and between day 3 and 10 % WL at 25 W (P<0.01). RQ at 50W was different between baseline and 1 week (P<0.001), 5 % WL (P<0.01), 10 % WL (P<0.01) and end (P<0.01).

4.7 Subjective feelings of Appetite

Changes in fasting subjective feelings of appetite can be seen in **Table 5**.

Table 5. Changes in subjective feelings of appetite in fasting over time.

	Baseline	Day 3	1 week	5 % WL	10 % WL	End	P-value
Hunger, cm	4.0±2.1	6.0±2.3	5.3±2.6	4.9±2.8	5.0±2.6	5.8±2.6	NSa)
Fullness, cm	2.9±1.9	2.2±1.4	3.4±2.2	3.9±2.1	2.8±1.9	2.9±2.4	NSa)
Desire, cm	4.8±2.6	6.4±2.0	5.8±2.8	5.8±1.7	6.3±2.0	6.2±2.4	NSa)
PFC*, cm	6.7±2.1	7.5±1.8	6.3±2.8	6.4±1.6	6.6±2.0	5.9±2.6	NSa)

Data presented as mean±SD. *PFC=Prospective food consumption. NS=Non significance. No significant main effect of time and gender was found on subjective feelings of appetite in fasting. **a)** Significance for main effect of time and gender.

No significant main effect of time, gender or time*gender interaction were found in subjective feelings of hunger, fullness, desire to eat and prospective food consumption in fasting. Post Hoc analysis revealed a trend towards an increase in subjective feelings of hunger from baseline to day 3 (*P*=0.058).

Changes in subjective feelings of appetite in the postprandial state (AUC) can be seen in **Figures 2 a-d5**.

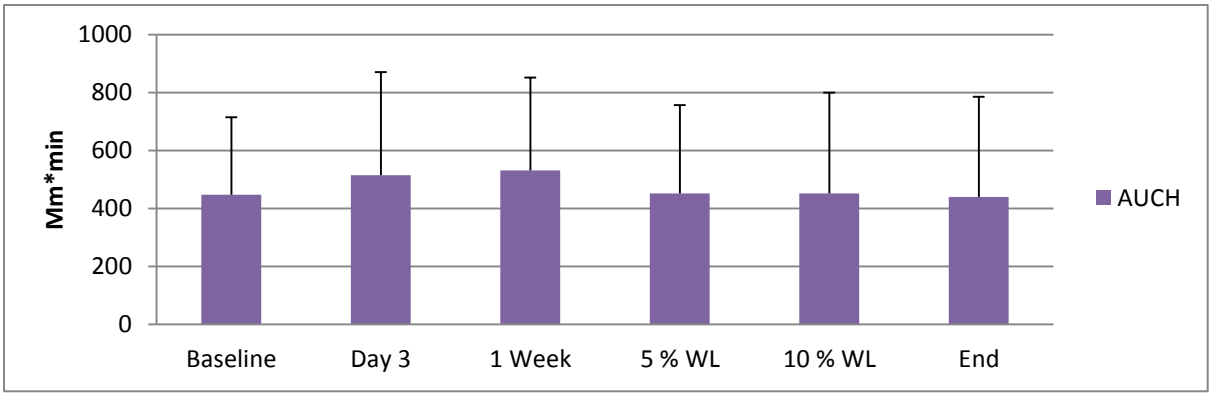

Figure 2a. AUC for subjective feelings of hunger (mean±SD) over time.

No significant main effect of time, gender or time*gender interaction were found in AUC for subjective feelings of hunger.

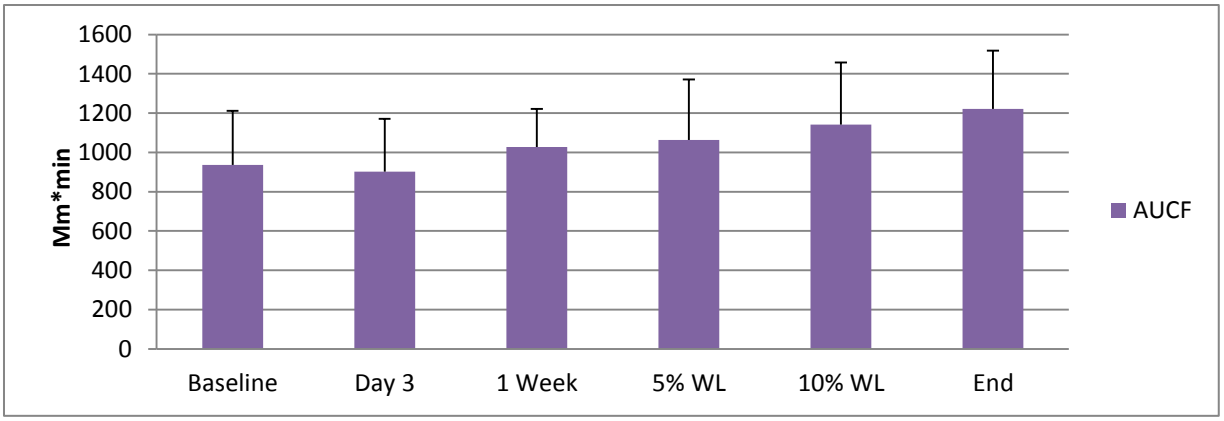

Figure 2b. AUC for subjective feelings of fullness (mean±SD) over time.

A significant main effect of time (P<0.01) and gender (P<0.01) was found in AUC for subjective feelings of fullness with women scoring higher than men (average difference of 298±88 mm*min), but no gender*time interactions. Further Post Hoc analysis revealed that there was a tendency towards a significant increase in AUC fullness between day 3 and end (P=0.070).

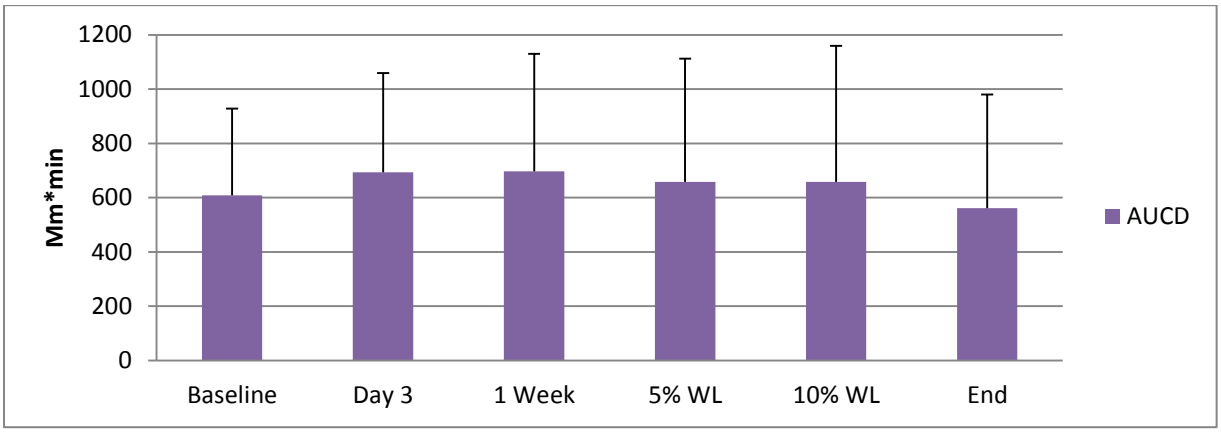

Figure 2c. AUC for subjective feelings of desire to eat (mean±SD) over time.

No significant main effect of time, gender or time*gender interaction (P<0.05) were found regarding AUC subjective feelings of desire to eat.

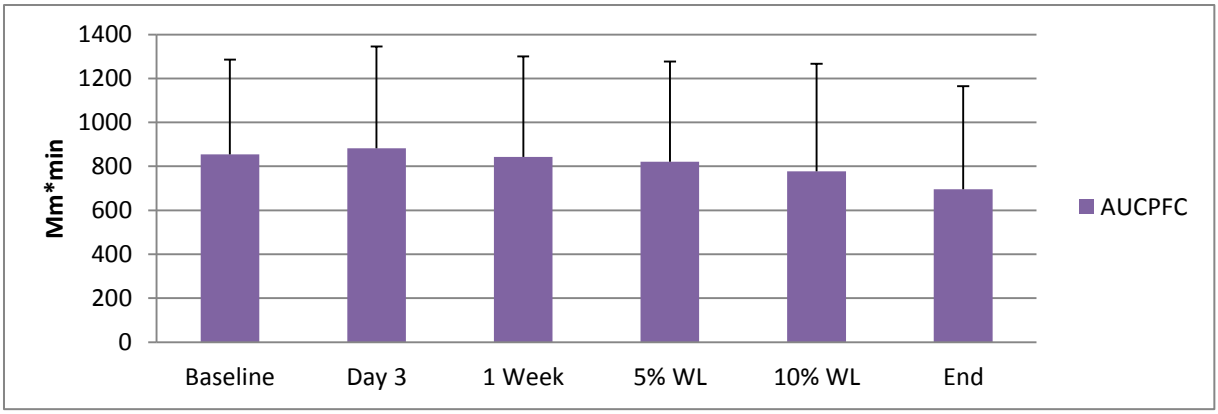

Figure 2d. AUC for subjective feelings of prospective food consumption (mean±SD) over time.

A significant main effect of time (P<0.05), but no main effect of gender or time*gender interaction, was found in AUC for subjective feelings of PFC. Further Post Hoc analysis revealed that there were a tendency towards a reduction in AUC for PFC between day 3 and end (P=0.071).

5. Discussion

This study aimed at identifying the timeline over each compensatory mechanisms, at level of both EI and EE, are activated during WL with an 8-week VLCD and to assess if gender modulates such compensatory mechanisms. A significant reduction in REE was first seen after 5 % WL with maintenance with progressive WL, and an increase in exercise efficiency at 10 W was only observed after 8 weeks on VLCD (average 16.4 % WL). A decrease in RQ with exercise was seen already at day 3, with a plateau at 5 % and 10 % WL at 10 and 25 W, respectively. RQ at rest declined, meaning that fat oxidation increased, throughout the 8 weeks of VLCD and significant changes were already seen at day 3 with further maintenance until end of study. On the other hand, subjective feelings of appetite did not change throughout the 8 weeks of VLCD, but a tendency towards increased subjective feelings of hunger in fasting was found at day 3. No significant differences between men and women were found in the activation of compensatory mechanisms.

A significant decline in REE (approximately 10 %) was observed first after 5 % WL (6.1±1.1 kg WL) and REE did not decline further with progressive WL. This suggests that a minimal WL of 5 % (6.1 kg) is needed to induce a significant reduction in REE and that after that threshold a larger WL will not induce further decline in REE with VLCD. Camps and colleagues (2013) reported a positive correlation between magnitude (9.6±4.1 kg) of WL and

reduction in REE after 8 weeks on VLCD (18). On the other hand, Doucet and colleagues (2001) found a significant decline in REE (6.4 % in men; 9.3 % in women) after only 2 weeks of energy restriction (-700 kcal/d) (41), despite a WL of only 1.5 kg in men and 0.9 kg in women. Even though in the present study participants achieved 5 % WL approximately at 2 weeks (day 14±4), a much larger WL (6.1±1.1 kg) and decline in REE was observed compared with Doucet and colleagues study (2001). Opposite to the findings in the present study, Kreitzman and colleagues (1992) reported a 14 % reduction in REE with 10 days of VLCD, with subsequent stability in spite of continuing WL during 10 weeks on VLCD (despite total WL of 16.2±2.4 kg) (51). The difference may be explained by 10 days of diet stabilization, with a fixed formula of 1600 kcal/d, before the start of VLCD in Kreitzman and colleagues (1992) study, that may have led to a faster decline in REE followed by a plateau. On the other hand, Foster and colleagues (1990) found approximately the same decrease in REE (17 %) after 8 weeks on VLCD as in the present study, but 4 weeks of refeeding (700-1200 kcal/d) increased REE to 8.4 % below initial REE despite weight stabilization (40). Furthermore, in the present study, men had a greater decrease in REE compared with women, but this is likely to be a result of a larger loss of FFM (kg) in men compared with women.

A significant increase in fat oxidation was seen after only 3 days of VLCD with significant WL (2.1 kg). However, most of the previous studies have found opposite findings, with a decrease in fat oxidation with diet-induced WL (WL of 10 kg or more) (12, 16, 25, 26, 52, 53), which again may contribute to difficulties in maintaining body weight after WL. However, studies showing decreased fat oxidation after WL have measured substrate oxidation in rest after 1-4 weeks of weight stabilization (12, 16, 25, 26, 52, 53), so the findings in the present study might be a result of the fact that the measurements were taken during active WL. On the other hand, an increase in fat oxidation was also found in formerly obese women after 53-days on VLCD with an average WL of 11.5 % (54), but those measurements were also taken during active WL. Furthermore, the significant reduction in RQ during exercise at 10 and 25 W, seen already at day 3 increased further at 5 % WL and 10 % WL, respectively, but was stabilized after that and until end of study. This suggest that the increase in fat oxidation with exercise reaches a plateau after 5 % WL (6.4 kg) at 10 W and 10 % WL (11.8 kg) at 25 W. Rosenbaum and colleagues (2013) also found a significant increase in RQ during exercise at 10 W with WL, but the increase was first seen after 10 % WL (10.2±5.4 kg) at the end of study (14). Furthermore, as mentioned earlier there was significant WL at all time points, but the decline in RQ could also be due to massive energy restriction,

and not only WL, that have resulted in increased levels of free fatty acids that may limit mitochondrial ATP and therefore reduce glucose oxidation (55).

An increase in exercise efficiency was only seen at 10 W at the end of the study (18.4 kg WL), which indicates that changes in exercise efficiency may only happen at very low intensity with a large amount of WL (>15 %). Rosenbaum and colleagues (2003) found increased exercise efficiency at 10 and 25 W after 10 % WL (14), however, this study did not use a VLCD, but participants received adjusted energy deficit during 5-8 weeks until they reached 10 % WL. On the other hand, Weinsier and colleagues (2000) reported no change in skeletal muscle work efficiency after 12.8 kg diet-induced WL at five different exercise tasks (56). This is unexpected given that the tasks reflected free-living PA as walking, cycling at 60 rpm, and stair climbing. However, the measurements by Weinsier and colleagues (2000) were done after 4 weeks of weight maintenance, while our measurements were performed immediately after the WL phase.

No significant changes in subjective feelings of appetite in fasting were found during or after 8 weeks on VLCD, but a tendency towards increased subjective feelings of hunger was seen at day 3 (2.1 kg WL) in fasting. These results can be due to the small sample size and multiple comparisons that make it difficult to find significant differences over time. On the other hand, ketosis could have prevented the increase in hunger and decrease in fullness usually seen with diet-induced WL, as previously shown by Sumithran and colleagues (2013). The authors reported no increase in hunger or reduction in fullness after 8 weeks of a ketogenic VLCD (57), despite an increase in fasting subjective feelings of hunger, desire to eat and PFC after 2 weeks of refeeding and WL maintenance. The same study had similar results as in this study, where participants ratings of subjective feelings of appetite, both in fasting and in the postprandial state, were unchanged after 8 weeks of VLCD (while participants were under ketosis) compared with baseline (57). On the other hand, Ratliff and colleagues (2009) also found that WL (6.7 kg) with ketosis was not associated with increased ghrelin levels or hunger feelings, but participants felt more satisfied, and wanted to eat less after the intervention (58). In the present study participants also reported a tendency to feel fuller and wanted to eat less between day 3 and end, when participants were under ketosis. Mars and colleagues (2006), on the other hand, found that energy restriction of 65 % increased hunger feelings after only 4 days with a significant WL (average 2.4 kg) (38). Therefore, WL without ketosis is likely to produce different outcomes compared with those reported in this study.

Strengths and limitations

This study has several strengths. First, compliance with the intervention was very good, based on both analysis of food diaries and measurement of ketone bodies. Second, "gold standard" techniques were used for the measurement of body composition (BodPod), REE (indirect calorimetry) and subjective feelings of appetite (VAS). Last, data was collected at several (six) time points over time allowing the evaluation of the effect of magnitude of WL on the activation of several compensatory mechanisms.

Unfortunately, this study also suffers from limitations. First, only 5 persons had valid data from the armbands, on all four time points, which may have distorted the findings regarding changes in PA over time. Second, the present study suffers from a small sample size. Time constrains and drop-outs prevented inclusion of more participants in the study. Even though the study had enough power to examine changes in REE it was probably underpowered to explore possible differences in appetite ratings and genders. Third, data should also have been collected when participants were weight stable and not in ketosis, to be able to evaluate the true effect of WL on compensatory mechanisms, particularly at the level of the appetite control system, since we know that ketosis (57, 58) modulate those mechanisms. Last but not least, the results from this study cannot be generalized to other diet-induced WL strategies, since we used a VLCD ketogenic diet. Different outcomes were likely to be found if a non-ketogenic diet had been used.

Practical implications of findings

The prevalence of obesity is growing and long-term WL maintenance remains the biggest challenge in obesity management, with the majority experiencing weight regain. Activation of REE after 5 % WL, in this study, indicates that a new change in EI and EE may be necessary for further WL when individuals have lost 5 % of initial weight with VLCD, but also moral support, which health professionals should be aware of when helping obese people to lose and sustain WL. Moreover, changes and plateaus in compensatory mechanisms were seen also at 10 % WL, which may also make this time point important for revising EI and EE again for further WL after ketogenic diets.

Future studies

As emphasized previously, more studies with larger sample sizes are needed and measurements should also be taken in energy balance, after a period of WL maintenance.

Furthermore, it would be interesting to assess the timeline over which compensatory mechanisms are activated with a non-ketogenic diet. Studies are also needed (with larger sample sizes) to look at gender differences in the timeline over which compensatory mechanisms are activated.

6. Conclusion

When losing weight with a VLCD (ketogenic diet), compensatory mechanisms seem to be activated at different time points. A significant reduction in REE was seen from 5 % WL with maintenance until end of study, while an increase in exercise efficiency at 10 W was only seen at the end of the intervention. Fat oxidation increased at day 3 with further maintenance with progressive WL, but fat oxidation during exercise increased and reached a plateau at 5 % and 10 % WL at 10 and 25 W, respectively. Participants subjective feelings of appetite did not change significantly over time, despite a tendency towards an increase in fasting hunger at day 3. Moreover, no gender differences were observed in compensatory mechanisms with WL. However, more studies with larger sample sizes are needed to further examine the timeline of activation of compensatory mechanisms with WL.

References

1. WHO. Obesity and overweight: World Health organization. http://www.who.int/mediacentre/factsheets/fs311/en/index.html.
2. Droyvold WB, Nilsen TI, Kruger O et al. Change in height, weight and body mass index: Longitudinal data from the HUNT Study in Norway. International journal of obesity (2005) 2006; 30: 935-9.
3. Hånes H, Graff-Iversen S, Meyer H. Fakta og helsestatistikk om overvekt og fedme hos voksne. www.fhi.no: Folhekseinstituttet, 2012. http://www.fhi.no/eway/default.aspx?pid=239&trg=List_6212&Main_6157=6263:0:25,6306&MainContent_6263=6464:0:25,6307&List_6212=6218:0:25,6317:1:0:0:::0:0.
4. Midthjell K, Lee CMY, Langhammer A et al. Trends in overweight and obesity over 22 years in a large adult population: the HUNT Study, Norway. Clinical Obesity 2013; 3: 12-20.
5. IOTF, EASO. Obesity in Europe: The case for action, 2002. http://www.iaso.org/site_media/uploads/Sep_2002_Obesity_in_Europe_Case_for_Action_2002.pdf.
6. Blackburn G. Effect of degree of weight loss on health benefits. Obes Res 1995; 3: 211-6.
7. Anderson JW, Konz EC, Frederich RC et al. Long-term weight-loss maintenance: a meta-analysis of US studies. Am J Clin Nutr 2001; 74: 579-84.
8. Kraschnewski JL, Boan J, Esposito J et al. Long-term weight loss maintenance in the United States. Int J Obes 2010; 43.
9. Wing RR, Hill JO. Successful Weight Loss Maintenance. Annu Rev Nutr 2001; 21: 323-41.
10. Rosenbaum M, Leibel RL. Adaptive thermogenesis in humans. International journal of obesity (2005) 2010; 34 Suppl 1: S47-55.
11. Cornier M-A. Is your brain to blame for weight regain? Physiology & Behavior 2011; 104: 608-12.
12. Ballor DL, Harvey-Berino JR, Ades PA et al. Decrease in fat oxidation following a meal in weight-reduced individuals: a possible mechanism for weight recidivism. Metabolism: clinical and experimental 1996; 45: 174-8.
13. Doucet E, Imbeault P, St-Pierre S et al. Greater than predicted decrease in energy expenditure during exercise after body weight loss in obese men. Clinical science (London, England : 1979) 2003; 105: 89-95.
14. Rosenbaum M, Vandenborne K, Goldsmith R et al. Effects of experimental weight perturbation on skeletal muscle work efficiency in human subjects. American journal of physiology Regulatory, integrative and comparative physiology 2003; 285: R183-92.
15. Rosenbaum M, Hirsch J, Gallagher DA et al. Long-term persistence of adaptive thermogenesis in subjects who have maintained a reduced body weight. Am J Clin Nutr 2008; 88: 906-12.
16. Leibel RL, Rosenbaum M, Hirsch J. Changes in Energy Expenditure Resulting from Altered Body Weight. New England Journal of Medicine 1995; 332: 621-8.
17. Van Gaal LF, Vansant GA, De Leeuw IH. Factors determining energy expenditure during very-low-calorie diets. The American journal of clinical nutrition 1992; 56: 224S-9S.
18. Camps SG, Verhoef SP, Westerterp KR. Weight loss, weight maintenance, and adaptive thermogenesis. The American journal of clinical nutrition 2013; 97: 990-4.
19. Camps SG, Verhoef SP, Westerterp KR. Weight loss-induced reduction in physical activity recovers during weight maintenance. The American journal of clinical nutrition 2013; 98: 917-23.
20. Redman LM, Heilbronn LK, Martin CK et al. Metabolic and behavioral compensations in response to caloric restriction: implications for the maintenance of weight loss. PLoS One 2009; 4: e4377.
21. Bonomi AG, Soenen S, Goris AH et al. Weight-Loss Induced Changes in Physical Activity and Activity Energy Expenditure in Overweight and Obese Subjects before and after Energy Restriction. PLoS One 2013; 8: e59641.

22. Foster GD, Wadden TA, Kendrick ZV et al. The energy cost of walking before and after significant weight loss. Medicine and science in sports and exercise 1995; 27: 888-94.
23. Poole DC, Henson LC. Effect of acute caloric restriction on work efficiency. The American journal of clinical nutrition 1988; 47: 15-8.
24. Weigle DS, Brunzell JD. Assessment of energy expenditure in ambulatory reduced-obese subjects by the techniques of weight stabilization and exogenous weight replacement. International journal of obesity 1990; 14 Suppl 1: 69-77;discussion -81.
25. Schutz Y, Tremblay A, Weinsier RL et al. Role of fat oxidation in the long-term stabilization of body weight in obese women. The American journal of clinical nutrition 1992; 55: 670-4.
26. Doucet E, St Pierre S, Almeras N et al. Changes in energy expenditure and substrate oxidation resulting from weight loss in obese men and women: is there an important contribution of leptin? The Journal of clinical endocrinology and metabolism 2000; 85: 1550-6.
27. Doucet E, Imbeault P, St-Pierre S et al. Appetite after weight loss by energy restriction and a low-fat diet-exercise follow-up. International journal of obesity and related metabolic disorders : journal of the International Association for the Study of Obesity 2000; 24: 906-14.
28. Sumithran P, Prendergast LA, Delbridge E et al. Long-term persistence of hormonal adaptations to weight loss. The New England journal of medicine 2011; 365: 1597-604.
29. Gilbert JA, Drapeau V, Astrup A et al. Relationship between diet-induced changes in body fat and appetite sensations in women. Appetite 2009; 52: 809-12.
30. Doucet E, St-Pierre S, Alméras N et al. Relation between appetite ratings before and after a standard meal and estimates of daily energy intake in obese and reduced obese individuals. Appetite 2003; 40: 137-43.
31. Romon M, Gomila S, Hincker P et al. Influence of Weight Loss on Plasma Ghrelin Responses to High-Fat and High-Carbohydrate Test Meals in Obese Woman. J Clin Endocrinol Metab 2006; 91: 1034-41.
32. Cummings DE, Weigle DS, Frayo RS et al. Plasma ghrelin levels after diet-induced weight loss or gastric bypass surgery. The New England Journal of Medicine 2002; 346: 1623-30.
33. Frübeck G, Rotellar F, Hernandez-Lizoain JL et al. Fasting Plasma Ghrelin Concentrations 6 Months after Gastric Bypass are not Determined by Weight Loss or Changes in Insulinemia. Obes Surg 2004; 14: 1208-15.
34. Adam TC, Jocken J, Westerterp-Plantenga MS. Decreased Glucagon-like Peptide 1 Release after Weight Loss in Overweight/Obese Subjects. Obes Res 2005; 13: 710-6.
35. Essah PA, Levy JR, Sistrun SN et al. Effect of weight loss by a low-fat diet and a low-carbohydrate diet on peptide YY levels. Int J Obes 2010; 34: 1239-42.
36. Pfluger PT, Kampe J, Castaneda TR et al. Effect of Human Body Weight Changes on Circulating Levels of Peptide YY and Peptide YY3–36. The Journal of Clinical Endocrinology & Metabolism 2007; 92: 583-8.
37. Pasiakos SM, Caruso CM, Kellogg MD et al. Appetite and endocrine regulators of energy balance after 2 days of energy restriction: insulin, leptin, ghrelin, and DHEA-S. Obesity (Silver Spring, Md) 2011; 19: 1124-30.
38. Mars M, de Graaf C, de Groot CP et al. Fasting leptin and appetite responses induced by a 4-day 65%-energy-restricted diet. International journal of obesity (2005) 2006; 30: 122-8.
39. Doucet E, Pomerleau M, Harper M. Fasting and Postprandial Total Ghrelin Remain Unchanged after Short-Term Energy Restriction. The Journal of Clinical Endocrinology & metabolism 2004; 89: 1727-32.
40. Foster GD, Wadde TA, Feurer ID et al. Controlled trial of the metabolic effects of a very-low-calorie diet: short-and-long term effects. The American Journal of Clinical Nutrition 1990; 51: 167-72.
41. Doucet E, St-Pierre S, Almeras N et al. Evidence for the existence of adaptive thermogenesis during weight loss. The British journal of nutrition 2001; 85: 715-23.
42. Henry CJ, Lightowler HJ, Marchini J. Intra-individual variation in resting metabolic rate during the menstrual cycle. The British journal of nutrition 2003; 89: 811-7.

43. Curtis V, Henry CJ, Ghusain-Choueiri A. Basal metabolic rate of women on the contraceptive pill. European journal of clinical nutrition 1996; 50: 319-22.
44. Svendsen PF, Jensen FK, Holst JJ et al. The effect of a very low calorie diet on insulin sensitivity, beta cell function, insulin clearance, incretin hormone secretion, androgen levels and body composition in obese young women. Scand J Clin Lab Invest 2012; 72: 410-9.
45. Hall KD, Sacks G, Chandramohan D et al. Quantification of the effect of energy imbalance on bodyweight. Lancet 2011; 378: 826-37.
46. Gradaschi R, Camerini G, Carlini F et al. Physical Activity After Surgically Obtained Weight Loss: Study with a SenseWear Armband in Subjects Undergoing Biliopancreatic Diversion. OBES SURG 2013: 1-6.
47. Bond DS, Unick JL, Jakicic JM et al. Objective assessment of time spent being sedentary in bariatric surgery candidates. Obesity surgery 2011; 21: 811-4.
48. Unick JL, Bond DS, Jakicic JM et al. Comparison of two objective monitors for assessing physical activity and sedentary behaviors in bariatric surgery patients. Obesity surgery 2012; 22: 347-52.
49. Compher C, Frankenfield D, Keim N et al. Best practice methods to apply to measurement of resting metabolic rate in adults: a systematic review. Journal of the American Dietetic Association 2006; 106: 881-903.
50. Stubbs RJ, Hughes DA, Johnstone AM et al. The use of visual analogue scales to assess motivation to eat in human subjects: a review of their reliability and validity with an evaluation of new hand-held computerized systems for temporal tracking of appetite ratings. The British journal of nutrition 2000; 84: 405-15.
51. Kreitzman SN, Coxon AY, Johnson PG et al. Dependence of weight loss during very-low-calorie diets on total energy expenditure rather than on resting metabolic rate, which is associated with fat-free mass. Am J Clin Nutr 1992; 56: 258S-61S.
52. Franssila-Kallunki A, Rissanen A, Ekstrand A et al. Effects of weight loss on substrate oxidation, energy expenditure, and insulin sensitivity in obese individuals. The American journal of clinical nutrition 1992; 55: 356-61.
53. van Aggel-Leijssen DP, Saris WH, Hul GB et al. Short-term effects of weight loss with or without low-intensity exercise training on fat metabolism in obese men. The American journal of clinical nutrition 2001; 73: 523-31.
54. Rabol R, Svendsen PF, Skovbro M et al. Reduced skeletal muscle mitochondrial respiration and improved glucose metabolism in nondiabetic obese women during a very low calorie dietary intervention leading to rapid weight loss. Metabolism: clinical and experimental 2009; 58: 1145-52.
55. Brehm A, Krssak M, Schmid AI et al. Increased lipid availability impairs insulin-stimulated ATP synthesis in human skeletal muscle. Diabetes 2006; 55: 136-40.
56. Weinsier RL, Hunter GR, Zuckerman PA et al. Energy expenditure and free-living physical activity in black and white women: comparison before and after weight loss. The American journal of clinical nutrition 2000; 71: 1138-46.
57. Sumithran P, Prendergast LA, Delbridge E et al. Ketosis and appetite-mediating nutrients and hormones after weight loss. European journal of clinical nutrition 2013; 67: 759-64.
58. Ratliff J, Mutungi G, Puglisi MJ et al. Carbohydrate restriction (with or without additional dietary cholesterol provided by eggs) reduces insulin resistance and plasma leptin without modifying appetite hormones in adult men. Nutrition research (New York, NY) 2009; 29: 262-8.

Appendix A - Consent form 1

Forespørsel om deltakelse i forskningsprosjektet
Hvordan holde vekten etter diettindusert vekttap?

Bakgrunn og hensikt

Dette er et spørsmål til deg om å delta i en forskningsstudie med utgangspunkt i en 8-ukers streng diett etterfulgt av ett års oppfølging med sikte på å stabilisere vekten. Forskerne har to hovedfokus i denne studien:

- Er de ulike oppfølgingsprogrammene like gode?
- Hvordan påvirkes hormonene som regulerer appetitt i diettens aktive fase?

Det er St. Olavs Hospital som er ansvarlig for studien.

Hva innebærer studien?

Studien er delt i to faser. Den første fasen er en 8-ukers diettperioden som vil være den samme for alle som deltar. En slik lavkalorikur kan gi noen bivirkninger (beskrevet senere). Når dietten er overstått vil du gå over i studiens andre fase som dreier seg om oppfølging med sikte på å opprettholde vekttapet.

Halvparten av pasientene vil få oppfølging i poliklinikken, mens den andre halvparten får oppfølging ved Røros Rehabilitering. Hvilken oppfølging du får er avhengig av hvor det er kapasitet for oppfølging på tidspunktet du inkluderes i studien. Oppfølgingen varer i ett år og du kan lese mer om den på neste side.

Undersøkelsene er stort sett de samme uansett hvilket oppfølgingsprogram du følger og innebærer for de aller fleste blodprøver, blodtrykk, målinger av energibehov, kroppsmasse og oksygenopptak, samt ulike former for spørreskjema.

Mulige fordeler, ulemper og bivirkninger
Fordelen med studiedeltakelse kan være at man går ned i vekt og oppnår bedre helse uten kirurgisk behandling. Deltakelse kan også gjøre at du blir bedre kjent med mekanismene i din egen kropp som påvirker appetitten. Dessuten vil du spare kostnader til mat i studiens diettfase (diettproduktene får du gratis ved sykehuset). Behandlingen anses ikke som risikabel. Undersøkelsene innebærer noen blodprøver.

Lavkalorikurer kan ha flere bivirkninger. Omfanget av disse varierer fra person til person og kan være enten helt fraværende eller temmelig plagsomme. Bivirkninger er forbigående. Rapporterte bivirkninger er:

- slapphet
- svimmelhet
- forstoppelse
- hårtap
- tørr hud
- neglene kan bli sprøere
- kvalme
- diaré
- forstyrret menstruasjonssyklus
- økt kuldefornemmelse

Hva skjer med prøvene og informasjonen om deg?
Prøvene tatt av deg og informasjonen som registreres om deg skal kun brukes slik som beskrevet i hensikten med studien. Alle opplysningene og prøvene vil bli behandlet uten navn og fødselsnummer eller andre direkte gjenkjennende opplysninger. En kode knytter deg til dine opplysninger og prøver gjennom en navneliste. Noen helseopplysninger vil også lagres i din pasientjournal, og disse vil være knyttet til ditt personnummer.

Det er kun autorisert personell knyttet til prosjektet som har adgang til navnelisten og som kan finne tilbake til deg. Det vil ikke være mulig å identifisere deg i resultatene av studien når disse publiseres.

Frivillig deltakelse
Det er frivillig å delta i studien. Du kan når som helst og uten å oppgi noen grunn trekke ditt samtykke til å delta i studien. Dette vil ikke få konsekvenser for din videre behandling. Dersom du ønsker å delta, undertegner du samtykkeerklæringen på siste side. Om du nå sier ja til å delta, kan du senere trekke tilbake ditt samtykke uten at det påvirker din øvrige behandling. Dersom du senere ønsker å trekke deg eller har spørsmål til studien, kan du kontakte studiekoordinator Hege Bjøru, telefon 40 87 34 24.

Studien er godkjent av Regional komité for medisinsk og helsefaglig forskningsetikk REK Midt-Norge.

Ytterligere informasjon om studien finnes i kapittel *A* – *utdypende forklaring av hva studien innebærer.*
Ytterligere informasjon om personvern og forsikring finnes i kapittel B – *Personvern, biobank, økonomi og forsikring.*

Samtykkeerklæring følger etter kapittel

Kapittel A – Utdypende forklaring av hva studien innebærer

Kriterier for deltakelse
De som kan delta i denne studien må

1. ha BMI mellom 35 og 45 kg/m^2,
2. være mellom 18 og 65 år,
3. ha et ønske om å gå ned i vekt ved hjelp av diett,
4. være relativt vektstabil siste tre måneder

Kvinner må dessuten enten være over menstruerende alder eller benytte p-piller.

Mange kan ha forsøkt dietter tidligere og du bør derfor tenke deg godt om hvorvidt dette er en behandling som er verdt å forsøke igjen. Hvis dette føles galt, så bør du ikke ta del i studien.

Bakgrunn for studien
Lavkaloridietter (< 800 kcal/dag) er en relativt sikker metode for å gå ned i vekt og gir også et raskt vekttap. Slike dietter kan gi vekttap i størrelse 10-15 % og med det også bedring i overvektsrelaterte sykdommer og risikofaktorer. Langtidseffektene er imidlertid usikre og særlige utfordringer er knyttet til opprettholdelse av vekttap på sikt. Det er behov for mer kunnskap om diettens vedlikeholdsfase, spesielt knyttet til tidspunktet man går over fra diettprodukter til mer normal, energiredusert kost.

Hovedhensikt med denne studien er å sammenligne opprettholdelse av vekttap etter 8-ukers lavkaloridiett hos pasienter som deltar i to ulike oppfølgingsprogram. Oppfølgingen varer i ett år.

Vi vil også se nærmere på hvordan den hormonelle appetittreguleringen endres i diettens aktive fase. Appetitten er et komplisert samspill av blant annet hormoner som både stimulerer og reduserer matlysten og vi vil følge utviklingen i disse i løpet av de ukene dietten varer. Det er hittil gjort lite forskning på dette.

Undersøkelser
Som del av studien vil du gjennomgå ulike undersøkelser.

- Veiing og kroppsmassemåling
- Blodtrykksmåling
- Blodprøver
    - Måling av appetitthormoner
    - Testing for kjente gener som disponerer for fedme
- Indirekte kalorimetri (måling av energibehov)
- Måling av oksygenopptak
- Spørreskjema

Undersøkelsene finner sted ved studiens start, ved avslutning av dietten, og ved avslutning av oppfølgingen (etter ett år).

Tidsskjema for diettperioden (8 uker) - felles for alle
Du vil få utdelt et variert utvalg av diettprodukter (milkshakes, smoothies, supper) tilsvarende et daglig energiinntak på 550 kcal (kvinner) og 660 kcal (menn). Du skal kun spise dette i diettens aktive fase (standardisert for alle), men du oppfordres til å drikke rikelig (minst 2,5

liter) vann og evt kalorifri drikke i tillegg. Du vil så få time hos en sykepleier i Obesitaspoliklinikken i studieuke 1, 2, 4, 6, og 8. Kostdagbok, veiing og urinprøver er del av denne fasen og bivirkninger rapporteres systematisk. I studieuke 8 får du time hos klinisk ernæringsfysiolog som vil foreskrive en ny diett av normalkost som du skal følge i året som kommer.

Tidsskjema for deltakere ved Røros Rehabilitering (1 år) - halvparten av deltakerne
For de som trekkes ut til å delta på Røros Rehabilitering, innebærer deltakelse tre opphold ved Røros. Hvert opphold varer i tre uker og gjøres unna i løpet av ett år. Oppholdene innebærer mye fysisk aktivitet, oppfølging av helsepersonell både individuelt og i grupper, samt matlaging i fellesskap. Mer informasjon og tidsplan for oppholdene vil bli distribuert senere.

Tidsskjema for deltakere ved Obesitaspoliklinikken (1 år) - halvparten av deltakerne
For de som trekkes ut til å delta i Obesitaspoliklinikkens program, innebærer det en individuell konsultasjon hos klinisk ernæringsfysiolog og senere gruppemøter med ulike helsepersonell. Gruppemøtene finner sted 3, 6, 9 og 12 mnd etter dietten og fokuserer mye på ernæring og fysisk aktivitet.

Studiedeltakerens ansvar
Det er studiedeltakerens ansvar å møte til avtalt tid. For de som deltar ved Røros Rehabilitering, må de påregne å være der gjennom hele treukersperiodene.

Kompensasjon og egenandel
Det gies ingen premiering for å delta i studien, men du vil få diettproduktene i diettens aktive fase gratis. Det er viktig å standardisere dietten slik at alle spiser det samme.

For deltakere ved Røros Rehabilitering vil fastlegen gi sykmelding for perioden oppholdene varer. NAV innvilger i de aller fleste kommunenes tilfelle også fritak for arbeidsgiverperioden, men det er også noen kommuner som ikke gjør dette pr i dag.

For deltakere ved Røros Rehabilitering vil det også tilkomme egenandel. Denne dekker behandling, kost og losji og betales inntil man når beløpsgrensen for Frikort 2. (Beløpsgrense fastsettes av myndighetene fra år til år.)

Kapittel B – Personvern, biobank, økonomi og forsikring

Personvern
Ulike opplysninger vil registreres om deg som del av dette prosjektet. Prøvesvar og innledende screeningnotat vil legges i din pasientjournal og er derfor personidentifiserbart. Opplysninger på bakgrunn av testene du gjennomgår og intervjuet vil lagres på sykehusets server og vil være avidentifiserte så lenge studien pågår (det vil si at et unikt ID-nummer erstatter navnet ditt). Kodenøkkelen som knytter navn til nummer makuleres når studien er slutt, slik at data da anonymiseres. Alle som jobber med data fra studien har taushetsplikt.

Vi vil benytte et internettbasert system for å samle spørreskjemadata. Dette betinger at du har tilgang til en datamaskin eller smartphone. Rapporteringssystemet krypterer dine svar slik at det ivaretar kravene til personvern.

St. Olavs Hospital ved administrerende direktør er databehandlingsansvarlig.

Biobank
Blodprøvene for analyser av appetitthormoner og mulige fedmegener som blir tatt vil bli lagret i en forskningsbiobank ved St. Olavs Hospital. Hvis du sier ja til å delta i studien, gir du også samtykke til at det biologiske materialet og analyseresultater inngår i biobanken. Overlege Bård Kulseng er ansvarshavende for forskningsbiobanken. Det biologiske materialet kan bare brukes etter godkjenning fra Regional komité for medisinsk og helsefaglig forskningsetikk (REK).

Rett til innsyn og sletting av opplysninger om deg og sletting av prøver
Hvis du sier ja til å delta i studien, har du rett til å få innsyn i hvilke opplysninger som er registrert om deg. Du har videre rett til å få korrigert eventuelle feil i de opplysningene vi har registrert. Dersom du trekker deg fra studien, kan du kreve å få slettet innsamlede prøver og opplysninger, med mindre opplysningene allerede er inngått i analyser eller brukt i vitenskapelige publikasjoner.

Økonomi
Studien finansieres over driften ved St. Olavs Hospital og Røros Rehabilitering. Diettproduktene for deltakerne er gitt av produsenten.

Forsikring
Studiedeltakerne omfattes av Norsk pasientskadeforsikring, jf. Pasientskadelovens §1.

Informasjon om utfallet av studien
Publikasjoner på bakgrunn av studien vil bli lagt ut på vår hjemmeside: www.stolav.no/overvekt.

Samtykke til deltakelse i studien

Jeg er villig til å delta i studien

(Signert av prosjektdeltaker, dato)

Jeg bekrefter å ha gitt informasjon om studien

(Signert, rolle i studien, dato)

Appendix B - Consent form 2

Forespørsel om deltagelse i *delstudie* knyttet til studien *Hvordan holde vekten etter diettindusert vekttap*

Bakgrunn og hensikt
Informasjonen du har lest for hovedstudien *Hvordan holde vekten etter diettindusert vekttap* gjelder også for denne studien. Bakgrunn for et eget informasjonsskriv for denne delstudien er at den også innebærer noen ekstra målinger/prøver. Prøvene er de samme som de som blir tatt i hovedstudien, men de tas litt oftere.

Bakgrunn for hyppigere prøver er ønsket om å tidfeste hvordan kroppens kompensatoriske mekanismer mot å gå ned i vekt opptrer. Med andre ord, på hvilket tidspunkt de setter inn sterkest, og når de eventuelt avtar.

Hva innebærer delstudien?
Delstudien innebærer ekstra prøver på dag 3 og dag 7 etter studiestart, samt i det du har tapt henholdsvis 5 og 10 % av vekten din. De to siste prøvene er derfor ikke mulig for oss å tidfeste på forhånd.

Dette er samme type prøver som vi ellers tar av deg i hovedstudien.

Frivillig deltagelse
Det er frivillig å delta i delstudien og du kan velge å si nei til denne delstudien, men samtidig være med i hovedstudien.

Samtykke til deltagelse i delstudien

Jeg er villig til å delta i studien

(Signert av prosjektdeltaker, dato)

Jeg bekrefter å ha gitt informasjonen om studien

(Signert, rolle i studien, dato)