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Magnitude of muscle wasting early after on-pump coronary artery bypass graft surgery and exploration of etiology

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Bullet points

°It remains uncertain whether significant fat-free mass wasting occurs early after CABG surgery, and the etiology of this wasting in this particular condition is unexplored.

°Significant fat-free mass wasting is present after CABG surgery, and this wasting effect is greater in younger patients and patients with greater increments in blood cortisol/testosterone ratios after surgery.

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Abstract

The magnitude and etiology of muscle wasting early after coronary artery bypass graft (CABG) surgery remains unknown. In the present study we assessed changes in fat-free mass early after CABG surgery and explored the possible etiology (relations with post-surgical changes in blood hormones, insulin resistance, subject characteristics, inflammation) for these changes. Fat-free mass was assessed before and 23 (range:25) days after CABG surgery in 25 subjects. Blood testosterone, cortisol, insulin-like growth factor-1 (IGF-1), growth hormone, sex-hormone binding globulin (SHBG), glucose, insulin, c-peptide, and c-reactive protein (CRP) concentrations were determined and free androgen index, cortisol/testosterone ratio and HOMA-IR index were all calculated before surgery, during the first three days after surgery, and at body composition re-assessment. Relationships between changes in fat-free mass and changes in blood parameters after surgery or subject characteristics were studied. After surgery, free androgen index and blood SHBG, testosterone and IGF-1 content decreased significantly, while HOMA-IR index, cortisol/testosterone ratio, blood growth hormone, insulin and CRP content increased significantly ($p<0.0025$, observed $\alpha>0.80$). Whole-body fat-free mass decreased significantly (by -1.9 (range:9.1)kg, $p<0.0025$, observed $\alpha=0.99$) after surgery. According to regression analysis, greater absolute fat-free mass loss was observed after CABG surgery in subjects who were younger, experienced a greater increase in blood cortisol/testosterone ratio after surgery, and/or underwent earlier body composition re-assessment ($p<0.05$). Significant decrements in fat-free mass were observed early after CABG surgery, especially in younger subjects and/or subjects with elevated blood cortisol/testosterone ratios after surgery. Interventions to preserve fat-free mass soon after CABG surgery are thus warranted.

Introduction

More than 200,000 coronary artery bypass graft (CABG) surgery procedures are carried out each year in the USA (Weiss and Elixhauser, 2014). Coronary artery bypass graft (CABG) surgery is a procedure employed to restore or optimise myocardial perfusion in coronary artery disease. In this surgical intervention, a venous or arterial graft is used to bypass the coronary occlusion or stenosis. Because cardioplegia (induction of temporary cardiac arrest) is often executed, the patient's circulation is coupled to a cardiopulmonary bypass and access to the heart is achieved by median sternotomy. After CABG surgery a 7-14 day hospital stay is generally required.

A likely consequence of CABG surgery is the considerable muscle wasting during the recovery period. This leads to a reduction in muscle strength, functional capacity and insulin sensitivity, and elevated mortality, especially in subjects aged >50 years (Szulc et al, 2010). Mechanistically, the loss of fat-free mass during recovery has been linked to a rapid decline in muscle protein synthesis rates (by ~36%) in the first four hours after CABG surgery (Caso et al, 2008). Muscle wasting should thus be considered as a significant adverse event after CABG surgery.

Whether muscle wasting actually occurs after CABG surgery remains however a topic of debate. Miller et al. (2007) reported no significant changes in whole-body fat-free mass within three months after CABG surgery. Conversely, Van Venrooij and colleagues (2012) reported a modest (0.8kg) but significant decrease in whole-body fat-free mass two months following CABG surgery. However, these studies were limited by the relatively late re-assessment of body composition after surgery. It could be argued that much of the reduction in fat-free mass was therefore not detected and/or the patients had already participated in rehabilitation interventions or engaged into regular (home-based) physical exercise allowing some restoration of fat-free mass (Marzolini et al, 2008). However, at present it is recommended to initiate exercise-based rehabilitation interventions (combination of endurance and resistance exercises) early after CABG surgery (immediately after hospital discharge) (Piepoli et al, 2012) which we and others routinely perform in clinical practice. Thus, it is necessary to clarify the extent of fat-free mass loss during recovery from CABG surgery. This is important

because recommendations on early tolerable muscle exercises are likely to emerge in case of acute fat-free mass wasting.

Moreover, associated factors with fat-free mass wasting after CABG surgery, if present, has not been explored in great detail. Previous studies have indicated that CABG surgery compromised endocrinologic function (alterations in the concentration of anabolic and catabolic hormones in the blood), lowered insulin sensitivity and generated an inflammatory reaction (Lehot et al, 1992; Roth-Isigkeit et al, 1997; Roth-Isigkeit et al, 2000; Henzen et al, 2003; Velissaris et al, 2004; Maggio et al, 2005). In addition, bed rest significantly lowered skeletal muscle protein synthesis rates, at least in healthy individuals (Kortebein et al, 2007). Such endocrinologic and metabolic changes, together with inflammation, bed rest and/or insulin resistance, thus may ultimately lead to reductions in fat-free mass. Moreover, pre-operative patient characteristics may determine changes in fat-free mass after CABG surgery. A deeper understanding of the mechanisms and/or patient characteristics leading to fat-free mass wasting after CABG surgery could lead to earlier preventive treatments and/or rehabilitation interventions.

In this study, the early impact of CABG surgery on fat-free mass was examined and the etiology of fat-free mass wasting after CABG surgery was explored. We hypothesized that significant fat-free mass wasting was present early after CABG surgery, and that such fat-free mass wasting was due to endocrine hormone disturbances, insulin resistance, inflammation, and/or subject characteristics.

Methods

Ethical approval

This study was approved by the local medical ethical committee (Jessa Hospital, Hasselt, Belgium), the study conformed to the standards set by the latest revision of the Declaration of Helsinki, and written informed consents were obtained from all subjects.

Subjects

After inviting 119 eligible Dutch speaking, Caucasian, candidates admitted to the hospital for elective CABG surgery (from March 2011 up to October 2013), 30 subjects agreed to participate in this study (Figure 1). The study sample size (n=30) was based on previous studies assessing the impact of CABG surgery on fat-free mass (Miller et al, 2007; van Venrooij et al, 2012). Due to post-operative complications and prolonged hospitalisation in five subjects, data from 25 subjects were analysed. As a result, data from the present study reflected changes in blood parameters and body composition after CABG surgery with a normal post-operative course. Subjects were excluded prior to study when (mild) pulmonary, neurologic, oncologic and/or nephrologic disease was present. Patients with metal implants or previous CABG or valve surgery were not included. Patients with previous percutaneous coronary intervention and/or myocardial infarction, but without heart failure, were allowed to this study.

Design

This was a prospective observational study. The day before CABG surgery body composition was assessed and a fasting blood sample was collected. At the first, second, and third day after CABG surgery, fasting blood samples were collected (for measurement of blood testosterone, cortisol, insulin-like growth factor 1, growth hormone, sex-hormone binding globulin, c-reactive protein, glucose, insulin and c-peptide content, free androgen index, cortisol/testosterone ratio, HOMA-IR index). At entry of rehabilitation (23 (range:25) days after surgery) body composition was re-assessed and a fasting blood sample was collected.

Primary outcome measures

The primary outcome measure was whole-body fat-free mass.

Secondary outcome measures

Secondary outcomes measures were blood testosterone, cortisol, insulin-like growth factor 1, growth hormone, sex-hormone binding globulin, c-reactive protein, glucose, insulin and c-peptide content, free androgen index, cortisol/testosterone ratio, HOMA-IR index, time to body composition re-assessment, hospitalisation and surgery duration and intubation time.

CABG surgery

On-pump CABG surgery through median sternotomy was performed on all subjects by one surgeon with similar methodology. During this procedure, patients were connected to an open circuit extracorporeal circulation (of cardio-pulmonary bypass), the aorta was cross-clamped and the heart was arrested by blood cardioplegia. Body temperature was lowered to 34°C. The average duration of the procedure (start median incision up to end of closing of sternotomy) was 227 (range:248) minutes. During hospital stay (10 (range:7) days) subjects received physical therapy (daily breathing exercises for a duration of 15 min/day and endurance exercises (walking and cycling against resistance, arm cranking without applied resistance) up to 30 min/day at a low intensity (exercise heart rate <120 bts/min).

Measurements

Body composition. Following assessment of body weight and length, segmental and whole-body adipose tissue mass and fat-free mass were determined in fed condition using whole-body dual x-ray absorptiometry (Glickman et al, 2004) (DXA; Lunar DPXL, Wisconsin, USA). From four subjects the composition of arm tissue could not be measured because they did not fit under the scanner. In

these subjects whole-body fat-free and adipose tissue mass was calculated by the sum of fat-free and adipose tissue mass of trunk and legs.

Blood parameters. Blood samples were collected between 07.00 and 09.00 AM after an overnight fast, and processed and stored at the University Biobank Limburg (UBiLim). Blood parameter analysis was performed by standard diagnostic tests at the clinical laboratory of the Jessa Hospital. Blood glycosylated hemoglobin content (HbA1c) was determined in whole EDTA blood via a commercial HPLC based method (Hi-Auto A1C Analyser, Menarini Diagnostics). Serum tubes were left to clot for 30 min and subsequently centrifuged for 5 min at 2500g. These serum samples were used to asses blood hormones affecting skeletal muscle protein synthesis such as total testosterone (by chemiluminescent ELISA on an automated ELISA device Architect i2000SR, Abbott), total cortisol (by direct chemiluminescence sandwich ELISA on ADVIA Centaur XP analyser, Siemens), total insulin-like growth factor 1 (IGF-1) (by ImmuneRadioMetric Assay [IMRA] IGF-I kit, Beckman Coulter) and total growth hormone (by hGH-IRMA kit, DiaSource), as well as for sex-hormone binding globulin (SHBG) (by 2-step Chemiflex ImmunoAssay on automated ELISA device Architect i2000SR, Abbott), insulin, c-peptide content (both by direct chemiluminescence sandwich ELISA on ADVIA Centaur XP, Siemens) and blood glucose level (by colorimetric Hexokinase - Glucose-6-fosfaat dehydrogenase method on a Beckman Coulter AU 2700 chemical analyser, Beckman Coulter). Lithium heparin plasma was obtained by spinning blood samples for 5 min at 2500g. In these samples the lipid profile (total cholesterol, triglycerides, low-density lipoprotein (LDL) cholesterol, high-density lipoprotein (HDL) cholesterol) was measured by enzymatic colorimetry, together with the inflammatory status based on blood c-reactive protein (CRP) content by antibody based turbidimetric method, on a Beckman Coulter AU 2700 chemical analyser. Homeostatic model assessment-insulin resistance (HOMA-IR) index was calculated to estimate whole-body insulin sensitivity: $[(\text{insulin}_{(mU/L)} * \text{glucose}_{(mmol/L)}]/22.5$ (Matthews et al, 1985). Free androgen index was calculated by: (blood total testosterone $\text{content}_{(nmol/l)} * 100$)/blood sex-hormone binding globulin $\text{content}_{(nmol/l)}$ (Ho et al, 2006). Catabolic-anabolic hormone balance was calculated by: blood total cortisol content/blood total testosterone content.

Statistical analysis

Statistical analyses were executed by using SPSS version 22.0. Shapiro-Wilk tests indicated that most data were not normally distributed. Therefore, data were expressed as median (range) and non-parametric tests were applied. Changes in blood parameters during follow-up were analysed by related-sample Friedman variance of ranks tests, in which Bonferroni corrections for multiple comparisons (pre-operative vs. post-operative, n=20) were applied (statistical significance was set at $p<0.0025$). Changes in body composition were analysed by Wilcoxon signed ranks tests, in which Bonferroni corrections for multiple comparisons (pre-operative vs. post-operative, n=20) were applied (statistical significance was set at $p<0.0025$). Next, univariate Spearman correlations were calculated, and a forward stepwise multivariate regression model was constructed to examine relations between changes in whole-body fat-free mass and baseline parameters (blood parameters, age, gender, baseline whole-body adipose and fat-free mass, intubation time, hospitalisation and surgery duration, time to re-assessment of body composition) or changes in blood parameters after surgery up to re-assessment of body composition (area under the curve minus pre-operative value) (Table 2). In final a multivariate linear regression model was created in which relations between changes in whole-body fat-free mass and detected significant independent predictors from the forward stepwise multivariate regression model were examined. In these regression models statistical significance was set at $p<0.05$. Observed statistical power was calculated by GPower v. 3.1.

Results

Subject characteristics

Twenty-five subjects were examined (2 females, median age 65 (33) years, Table 1). Three subjects were revascularised by CABG surgery after acute myocardial infarction and others for stable angina with a positive stress test. Subjects were intubated for 10 (44) hours after CABG surgery and were hospitalised for 10 (7) days. Following medications were taken before surgery: anticoagulants (n=23), nitrates (n=11), beta-blockers (n=15), statins (n=19), calcium antagonists (n=3), metformin (n=5), sulfonylurea (n=2), insulin (n=1), diuretics (n=3), angiotensin receptor blockers (n=1), ACE inhibitors (n=9). Following medications were taken after surgery (at day of body composition re-assessment): anticoagulants (n=24), nitrates (n=4), beta-blockers (n=24), statins (n=22), calcium antagonists (n=7), metformin (n=5), sulfonylurea (n=2), insulin (n=1), diuretics (n=5), angiotensin receptor blockers (n=1), ACE inhibitors (n=5).

Changes in blood parameters during follow-up

During the first three days after surgery a significant inflammatory reaction and insulin resistance occurred, as can be observed by significant increases in blood CRP content and HOMA-IR index ($p<0.0025$, Table 2). Significant decreases in blood testosterone, IGF-1, and SHBG content, free androgen index, and increases in blood growth hormone and insulin content and cortisol/testosterone ratio, were observed ($p<0.0025$, Table 2). All significant changes in blood parameters were of sufficient observed statistical power ($\alpha>0.80$). At 23 (25) days of follow-up, all blood parameters were normalised.

Changes in body composition during follow-up

Together with a decrease in whole-body adipose tissue mass (from 24.0 (44.1)kg to 22.1 (44.5)kg, decrease of -1.4 (4.0)kg, $p<0.0025$), trunk adipose tissue mass (by -1.1 (3.9)kg, $p<0.0025$) and leg adipose tissue mass (by –0.2 (3.6) kg, $p<0.0025$), whole-body fat-free mass decreased significantly

(from 54.1 (24.8)kg to 52.7 (25.3)kg, decrease of -1.9 (9.1)kg, observed α=0.99, p<0.0025) after CABG surgery (Figure 2). Furthermore, significant decreases in fat-free mass were observed in the arms (by -0.5 (0.8)kg, p<0.0025) and legs (change of -0.5 (4.8)kg, p<0.0025) after CABG surgery, but not in the trunk (change of -0.7 (7.1)kg, p>0.0025). A great heterogeneity in whole-body fat-free mass loss was observed (Figure 3). Relative changes in fat-free mass in the different body regions were as follows: whole-body (by -4.4 (15.3)%), trunk (by -2.8 (22.5)%), legs (by -2.8 (25.8)%), and arms (by -8.0 (8.2)%).

Correlations

Univariate correlations (p<0.05) were found between magnitude of whole-body fat-free mass change after surgery and time to body composition re-assessment (r=0.43), age (r=0.56), pre-operative blood c-peptide (r=-0.53) and cortisol (r=0.50) content, post-operative change in blood cortisol content (r=-0.50) and free androgen index (r=0.47) (Figure 4). A correlation was found between age and changes in HOMA-IR (r=-0.45, p<0.05) after CABG surgery, but not with changes in blood hormones or inflammation markers. Age did not correlate significantly with time to re-assessment of body composition (p>0.05). Baseline fat-free mass did not correlate with changes in fat-free mass after CABG surgery (p>0.05).

Regression analysis

Greater increases in blood cortisol/testosterone ratio after surgery (standardized coefficient β=0.64, p<0.001), younger age (standardized coefficient β=0.68, p<0.001), and earlier re-assessment of body composition (standardized coefficient β=0.34, p=0.028) were independently related to greater absolute whole-body fat-free mass loss after CABG surgery (model adjusted r^2= 0.75, p<0.001). Also a trend for a relation between amount of fat-free mass loss and duration of surgery was found (standardized coefficient β=0.28, p=0.052).

Discussion

In the present paper, we demonstrate that significant decrements in fat-free mass occurred within the first weeks after CABG surgery. Moreover, we also show that more severe absolute reductions in fat-free mass are experienced by younger subjects, subjects with greater increases in blood cortisol/testosterone ratio after surgery and/or earlier re-assessment of body composition.

According to our data, CABG patients are likely to enter early rehabilitation programmes (within a few weeks after surgery) with reduced fat-free mass. Especially in the arms a pronounced reduction in fat-free mass (by -8.1±2.1%) was noticed. The reduction in fat-free mass could however have been mitigated by the early in-hospital rehabilitation program after CABG surgery. So it may be speculated that even greater fat-free mass decrements may be present after CABG surgery when such early rehabilitation intervention is absent. A reduction in body weight after CABG surgery should thus not be considered as a reduction in adipose tissue mass only. Given that fat-free mass wasting is associated with the development of insulin resistance, lowered muscle strength and functional capacity, and elevated mortality (Szulc et al, 2010), attempts should be made to minimize such post-operative wasting in CABG patients. In this regard, exercise-based rehabilitation interventions should be implemented soon after CABG surgery. Such interventions should involve resistance exercises in preference to endurance exercises due to the former being more strongly associated with hypertrophy and increased strength (Marzolini et al, 2008). In these interventions, the arm muscles must be targeted specifically. To optimize medical safety of resistance exercise training of the upper extremities early after CABG surgery, it is important to avoid the Valsalva manoeuvre, to assure that the sternum is stable, and only execute bilateral exercises with equal weights (Piepoli et al. 2012). It may also be speculated that nutritional support, such as protein supplementation (preferentially used in combination with resistance exercise) would augment fat-free mass regain in CABG patients (Cermak et al, 2012). Future studies should explore whether such optimised early exercise training programmes could minimize or even prevent fat-free mass wasting after CABG surgery.

In previous studies only small changes in whole-body fat-free mass were observed after CABG surgery (Miller et al, 2007; van Venrooij et al, 2012). At first glance this may seem to contradict our finding of considerable fat-free mass loss. However, in the present study, a smaller decline in fat-free mass was observed in patients with a later re-examination of body composition after CABG surgery. In agreement with this finding Miller et al. (2007) did not observe changes in whole-body fat-free mass at three months after CABG surgery, while van Venrooij et al. (2012) found a small significant decrease in whole-body fat-free mass at two months after CABG surgery. It could be hypothesized that a spontaneous regain in fat-free mass occurs after CABG surgery. Moreover, it could also be proposed that patients engaged into rehabilitation, or home-based physical activities, within these two to three months after CABG surgery in previous studies, resulting in fat-free mass regain. In the present study, patients did not start their ambulatory rehabilitation programme before body composition re-assessment after surgery. However, we did not monitor home-based physical activity so it remains uncertain whether spontaneous fat-free mass regain occurs after CABG surgery or whether this regain is related to home-based physical activity.

Despite the significant fat-free mass wasting in the total subject cohort after CABG surgery, the severity of this wasting was considerably different between individuals. Some subjects experienced severe fat-free mass wasting (6.3kg decrease in fat-free mass) while others did not (2.7kg gain in fat-free mass). Based on data from the present study, and in agreement with data from previous studies (Lehot et al, 1992; Roth-Isigkeit et al, 1997; Roth-Isigkeit et al, 2000; Henzen et al, 2003; Velissaris et al, 2004; Maggio et al, 2005), CABG surgery led to systemic inflammation and a decrease in blood anabolic hormone content (insulin-like growth factor 1 (IGF-1) and testosterone), while blood catabolic hormone content (cortisol) remained stable (suggesting a predominantly catabolic hormonal milieu, based on cortisol/testosterone ratio). Moreover, subjects were intubated and subsequently hospitalized during 10 (7) days, which led to a reduced physical activity level. In addition, subjects developed insulin resistance (six patients were diagnosed with diabetes before CABG surgery); patients were in need of exogenous insulin administration (Actrapid®) to prevent

excessive hyperglycemia (which in part explains the elevated post-operative blood insulin levels). To explain the etiology for the heterogeneity in fat-free mass wasting after CABG surgery, we therefore examined which of those factors/changes and patient features were independently related to greater fat-free mass wasting after CABG surgery. From this examination, it was observed that more severe fat-free mass wasting was experienced by subjects with greater increases in blood cortisol/testosterone ratio after surgery and/or younger subjects. Because blood cortisol levels remained stable during the first days after CABG surgery, but blood testosterone levels decreased significantly, a catabolic hormonal environment developed. According to previous investigations, the relative predominance of blood cortisol (due to lowered blood testosterone content) could lead to fat-free mass wasting by a combination of lowered protein synthesis (due to lowered transport of amino acids into the muscle, inhibition of insulin and IGF-1 action, and/or inhibition of phosphorylation of eIF4E-binding protein-1 and ribosomal protein S6 kinase-1) and increased protein breakdown (due to activation of cellular proteolytic systems such as ubiquitin-proteasome system, lysosomal system (cathepsins) and calcium-dependent system (calpains)) (Schakman et al, 2008). In addition, lowered blood testosterone levels could further lead to fat-free mass wasting by an increase in muscle protein degradation due to lowered amino acid uptake into the cell, binding of this hormone to intracellular androgen receptors (causing less transcription of specific genes), decrease in IGF-1 production and/or less inhibition of cortisol signaling (Vingren et al, 2010). Future studies are warranted to verify whether these physiological changes are indeed instrumental to the fat-free mass wasting process after CABG surgery. Because of the predominant catabolic hormonal environment after CABG surgery and its relation with fat-free mass wasting, other laboratories have already generated initial data concerning the supplementation of testosterone after CABG surgery to prevent fat-free mass wasting (Maggio et al, 2012).

Younger subjects experienced greater fat-free mass loss after CABG surgery in the present study. The signalling pathways leading to muscle atrophy and amount of fat-free mass wasting seems significantly affected by age. For example, in young and old subjects, molecular signalling pathways

governing skeletal muscle protein synthesis decrease significantly as result of immobilisation (Suetta et al, 2012). However, in older subjects higher Akt phosphorylation and early up-regulation of IGF-1 Ea and mechano growth factor (MGF), in combination with a lower atrogin-1 and muscle-specific muscle ring finger 1 (MURF-1) mRNA expression, has been observed during immobilisation (Suetta et al, 2012). These adaptations collectively lead to less fat-free mass wasting as result of immobilisation in older patients (Suetta et al, 2012). However, this hypothesis remains to be verified in CABG patients. Because changes in blood hormones after surgery were not related to age in the present study, it is speculated that greater fat-free mass wasting in younger subjects is not due to different changes in blood hormones. It could be speculated that younger subjects had a greater pre-operative fat-free mass, and thus greater reductions in fat-free mass can be anticipated in these subjects. However, in this study pre-operative fat-free mass did not correlate ($p>0.05$) with magnitude of fat-free mass wasting after CABG surgery.

It could be argued that fat-free mass wasting after CABG surgery is mainly due to bed rest (whole-body muscle disuse). Indeed, previous studies have shown that a 7-to-14 day bed rest was associated with significant fat-free mass wasting (Ferrando et al, 1996; Drummond et al, 2012) and that the anabolic response to amino acid intake was significantly suppressed (Drummond et al, 2012). The magnitude of fat-free mass wasting in the present study seemed comparable to those previously published data (Ferrando et al, 1996; Drummond et al, 2012). Considering the lack of a control who underwent bed rest only in this study, data from the present study should be evaluated in light of this shortcoming. However, subjects from the present study executed endurance exercises on daily basis under supervision of a physical therapist, were required to sit upright as often as possible and to walk in a nearby corridor. Therefore, it is likely that substantially more physical activity was present in our study as opposed to those previous studies (where recruited healthy volunteers were not allowed to leave the bed under carefully controlled laboratory conditions). Therefore, the observed fat-free mass wasting in the present study is probably not completely explained by physical inactivity/bed rest alone. Unfortunately, physical activity was not monitored

during hospitalisation and at home in this study. Future studies should thus examine the impact of post-operative physical activity level on fat-free mass after CABG surgery.

In the present study CABG surgery led to some changes in blood hormones that could be of interest for further study or are clinically relevant. However, it has to be mentioned that the endocrinologic impact of CABG surgery remains a topic of intense debate and is complex. After CABG surgery hyperinsulinemia occurred. CABG surgery provokes an inflammatory response (characterised by increased blood c-reactive protein content) and a reduction in blood testosterone content. Moreover, patients were sedated for several hours so skeletal muscle activity was significantly suppressed. Although many more factors were involved, these changes would, at least in part, induce insulin resistance. To overcome this insulin resistance and avoid excessive hyperglycemia, exogenous insulin (Actrapid®) was administered to the patients. This explains why a state of hyperinsulinemia was present after CABG surgery while blood c-peptide levels (indicative of pancreatic activity) remained stable after surgery. Moreover, in presence of an insulin-resistant state the protein synthesis stimulating effect of insulin on muscle tissue could have been suppressed (Bassil and Gougeon, 2013). In addition, CABG surgery, or any other major surgical procedure, is associated with rapid elevations in blood growth hormone content (Wallin, 2007; Nakhjavani et al, 2009). Although the etiology for such post-operative growth hormone elevation remains speculative, our findings are in line with these observations. Moreover, patients who underwent CABG surgery seemed to develop growth hormone resistance for currently unknown reasons (Wallin, 2007; Nakhjavani et al, 2009). A consequence of such growth hormone resistance is the lowering in blood IGF-1 levels (Wallin, 2007; Nakhjavani et al, 2009), as was observed in the present study. A reduction in circulatory IGF-1 levels in turn lowers insulin sensitivity (Wallin, 2007).

In the present study, significant decrements in adipose tissue mass were observed after CABG surgery. These findings are in disagreement with the study of van Venrooij et al. (2012) were no changes in adipose tissue mass were observed after CABG surgery. The reason for this discrepancy in outcomes between studies remains highly speculative. Data on the impact of CABG surgery on

adipose tissue mass are however scarce but can be clinically relevant due to the relation between (changes in) adipose tissue mass and clinical outcomes. It thus follows that this topic should deserve greater attention in the near future. In the present study, explaining the etiology of adipose tissue mass loss after CABG surgery is difficult due to a lack of detailed recordings of caloric and macronutrient intakes during hospitalisation.

This study may be limited by the small sample size. However, significant changes in body composition are observed after CABG surgery ($p<0.0025$) and changes in whole-body fat-free mass are of sufficient statistical power ($\alpha>0.80$). When using regression analyses to detect independent predictors of fat-free mass wasting after CABG surgery, findings are more robust when examining a large number of subjects. When using DEXA scans it is assumed that fat-free tissue hydration status is unaltered during follow-up. This cannot be confirmed in the present study. Subjects were predominantly male (92%). However, females were as frequently requested to participate in this study as males.

As a result of CABG surgery, significant fat-free mass wasting is observed and more severe wasting is experienced by subjects who are younger and/or with a greater increase in blood cortisol/testosterone ratio after surgery. Interventions for preservation of fat-free mass should thus be implemented or studied early after the execution of CABG surgery.

References

Bassil MS, & Gougeon R (2013). Muscle protein anabolism in type 2 diabetes. *Curr Opin Clin Nutr Metab Care* 16, 83-88.

Caso G, Vosswinkel JA, Garlick PJ, Barry MK, Bilfinger TV, & McNurlan MA (2008). Altered protein metabolism following coronary artery bypass graft (CABG) surgery. *Clin Sci* 114, 339-346.

Cermak N, Res PT, de Groot LC, Saris WH, & van Loon LJ (2012). Protein supplementation augments the adaptive response of skeletal muscle to resistance-type exercise training: a meta-analysis. *Am J Clin Nutr* 96, 1454-1464.

Drummond MJ, Dickinson JM, Fry CS, Walker DK, Gundermann DM, Reidy PT, et al (2012). Bed rest impairs skeletal muscle amino acid transporter expression, mTORC1 signaling, and protein synthesis in response to essential amino acids in older adults. *Am J Physiol Endocrinol Metab* 302, E1113-1122.

Ferrando AA, Lane HW, Stuart CA, Davis-Street J, & Wolfe RR (1996). Prolonged bed rest decreases skeletal muscle and whole body protein synthesis. *Am J Physiol* 270, E627-633.

Glickman SG, Marn CS, Supiano MA, & Dengel DR (2004). Validity and reliability of dual-energy X-ray absorptiometry for the assessment of abdominal adiposity. *J Appl Physiol* 97, 509-514.

Henzen C, Kobza R, Schwaller-Protzmann B, Stulz P, & Briner VA (2003). Adrenal function during coronary artery bypass grafting. *Eur J Endocrinol* 148, 663-668.

Ho CKM, Stoddart M, Walton M, Anderson RA, & Beckett GJ (2006). Calculated free testosterone in men: comparison of four equations and with free androgen index. *Ann Clin Biochem* 43, 389-397.

Kortebein P, Ferrando A, Lombeida J, Wolfe R, & Evans WJ (2007). Effect of 10 days of bed rest on skeletal muscle in healthy older adults. *JAMA* 297, 1772-1774.

Lehot JJ, Piriz H, Villard J, Cohen R, & Guidollet J (1992). Glucose homeostasis: comparison between hypothermic and normothermic cardiopulmonary bypass. *Chest* 102, 106-111.

Maggio M, Ceda GP, De Cicco G, Cattadori E, Visioli S, Ablondi F, et al (2005). Acute changes in circulating hormones in older patients with impaired ventricular function undergoing on-pump coronary artery bypass grafting. *J Endocrinol Invest* 28, 711-719.

Maggio M, Nicolini F, Cattabiani C, Beghi C, Gherli T, Schwartz RS, et al (2012). Effects of testosterone supplementation on clinical and rehabilitative outcomes in older men undergoing on-pump CABG. *Contemp Clin Trials* 33, 730-738.

Marzolini S, Oh PI, Thomas SG, & Goodman JM (2008). Aerobic and resistance training in coronary disease: single versus multiple sets. *Med Sci Sports Exerc* 40, 1557-1564.

Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, & Turner RC (1985). Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia* 28, 412-419.

Miller LE, Pierson LM, Pierson ME, Kiebzak GM, Ramp WK, Herbert WG, et al (2007). Changes in bone mineral and body composition following coronary artery bypass grafting in men. *Am J Cardiol* 99, 585-587.

Nakhjavani M, Esteghamati A, Hamidi S, Esfahanian F, Nabavi H, Abbasi, M et al (2009). Changes in growth hormone and insulin-like growth factor-I levels in the acute stage after open heart surgery and at the time of discharge. *Exp Clin Endocrinol Diabetes* 117, 413-416.

Piepoli MF, Corrà U, Benzer W, Bjarnason-Wehrens B, Dendale P, Gaita D, et al (2012). Cardiac Rehabilitation Section of the European Association of Cardiovascular Prevention and Rehabilitation. Secondary prevention through cardiac rehabilitation: from knowledge to implementation. A position paper from the Cardiac Rehabilitation Section of the European Association of Cardiovascular Prevention and Rehabilitation. *Eur J Cardiovasc Prev Rehabil* 17, 1-17.

Roth-Isigkeit A, Dibbelt L, Schmucker P, & Seyfarth M (2000). The immune-endocrine interaction varies with the duration of the inflammatory process in cardiac surgery patients. *J Neuroendocrinology* 12, 546-552.

Roth-Isigkeit AK, & Schmucker P (1997). Postoperative dissociation of blood levels of cortisol and adrenocorticotropin after coronary artery bypass grafting surgery. *Steroids* 62, 695-699.

Schakman O, Gilson H, & Thissen JP (2008). Mechanisms of glucocorticoid-induced myopathy. *J Endocrinol* 197, 1-10.

Suetta C, Frandsen U, Jensen L, Jensen MM, Jespersen JG, Hvid LG, et al (2012). Aging affects the transcriptional regulation of human skeletal muscle disuse atrophy. *Plos One* 7, e51238.

Szulc P, Munoz F, Marchand F, Chapurlat R, & Delmas PD (2010). Rapid loss of appendicular skeletal muscle mass is associated with higher all-cause mortality in older men: the prospective MINOS study. *Am J Clin Nutr* 91, 1227-1236.

van Venrooij LM, Verberne HJ, de Vos R, Borgmeijer-Hoelen MM, van Leeuwen PA, & de Mol BA (2012). Postoperative loss of skeletal muscle mass, complications and quality of life in patients undergoing cardiac surgery. *Nutrition* 28, 40-45.

Velissaris T, Tang AT, Murray M, Mehta RL, Wood PJ, Hett DA, et al (2004). A prospective randomized study to evaluate stress response during beating-heart and conventional coronary revascularization. *Ann Thorac Surg* 78, 506-512.

Vingren JL, Kraemer WJ, Ratamess NA, Anderson JM, Volek JS, & Maresh CM (2010). Testosterone physiology in resistance exercise and training: the up-stream regulatory elements. *Sports Med* 40, 1037-1053.

Wallin M (2007). Doctoral thesis: *The GH/IGF-1 system during surgery and catabolism – focus on metabolism and heart function*. P. 1-9, Karolinska Institutet.

Weiss AJ & Elixhauser A (2014). Trends in Operating Room Procedures in U.S. Hospitals, 2001–2011. In: *Healthcare Cost and Utilization Project (HCUP) Statistical Briefs.* Rockville (MD): Agency for Health Care Policy and Research (US), 2006-14.

Additional information

Competing interests

Dominique Hansen, Loes Linsen, Kenneth Verboven, Marc Hendrikx, Jean-Luc Rummens, Monique van Erum, Bert O Eijnde and Paul Dendale declare that they have no conflict of interest.

Author contributions

Conception and design of experiments: Dominique Hansen, Loes Linsen, Monique van Erum, Bert O Eijnde, Paul Dendale.

Collection, analysis and interpretation of data: Dominique Hansen, Loes Linsen, Kenneth Verboven, Jean-Luc Rummens, Marc Hendrikx, Paul Dendale.

Drafting the article or revising it critically for important intellectual content: Dominique Hansen, Loes Linsen, Kenneth Verboven, Marc Hendrikx, Monique van Erum, Bert O Eijnde, Paul Dendale.

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Table 1 Subject characteristics

Age (yrs)	65 (33)
Gender (n females)	2
Body mass index (kg/m²)	27.4 (19.8)
Cardiovascular risk factors before surgery	
Obesity (body mass index ≥30kg/m², n)	6
Diabetes (blood $HbA1_c$ ≥6.5% or taking medication, n)	6
Hypertension (blood pressure ≥140/90mmHg or taking medication, n)	16
Hypercholesterolemia (blood cholesterol level ≥200mg/dl or taking medication, n)	19
Active smoking (n)	6
Revascularised coronary arteries	
Left descending artery (n)	25
Right coronary artery (n)	17
Circumflex artery (n)	15
Acute myocardial infarction (n)	3
Duration of intubation (hours)	10 (44)
Duration of hospitalisation (days)	10 (7)
Interval between pre-operative vs post-operative DEXA scan (days)	23 (25)
Blood glycosylated hemoglobin content (%)	5.7 (2.9)
Blood total cholesterol content (mg/dl)	145 (142)
Blood LDL cholesterol content (mg/dl)	76 (116)
Blood HDL cholesterol content (mg/dl)	35 (47)
Blood triglyceride content (mg/dl)	126 (302)

Data are expressed at median (range).

Abbreviations: HDL, high-density lipoprotein; LDL, low-density lipoprotein.

Table 2 Baseline blood parameters and impact of CABG surgery

	pre-operative	day 1 after CABG	day 2 after CABG	day 3 after CABG	23 (25) days after CABG
glucose (mmol/l)	5.3 (11.1)	6.7 (5.0)	6.8 (6.4)	6.5 (10.4)	5.7 (6.6)
c-peptide (ng/ml)	1.6 (4.2)	0.7 (3.4)	0.9 (6.1)	1.7 (12.2)	1.7 (2.5)
insulin (mU/l)	8.2 (29.1)[a]	35 (167)	34 (77)	30 (174)	10 (23)
HOMA-IR index	2.1 (7.6)[a]	9.0 (53)	7.7 (26.7)	7.4 (83.0)	2.9 (5.7)
c-reactive protein (mg/dl)	0.1 (2.9)[a]	9.6 (11.4)	21 (25	22.9 (28.9)	0.5 (9.4)
cortisol (total, µg/dl)	17 (23)	21 (23)	17 (19)	18 (23)	16 (15)
testosterone (total, ng/ml)	3.6 (7.0)[a]	0.7 (2.0)	0.7 (2.0)	0.8 (2.6)	3.1 (9.0)
sex-hormone binding globulin (nmol/l)	38 (58)[a]	25 (49)	24 (39)	28 (32)	38 (89)
insulin-like growth factor 1 (µg/l)	142 (183)[a]	105 (103)	103 (77)	83 (85)	149 (175)
growth hormone (µg/l)	0.1 (0.5)[a]	1.2 (3.7)	0.8 (2.5)	0.4 (1.4)	0.5 (3.6)
free androgen index (%)	30 (65)[a]	11 (24)	8 (25)	9 (26)	28 (98)
cortisol/testosterone ratio	5 (210)[a]	25 (604)	23 (169)	22 (105)	6 (142)

[a]Significantly different as opposed to post-operative values in the first three days only ($p<0.0025$).

Data are expressed as median (range).

Abbreviations: HOMA-IR, homeostatic model assessment-insulin resistance.

Figure 1

The study flowchart is depicted in Figure 1. After inviting 119 eligible candidates admitted to the hospital for elective CABG surgery, 30 subjects agreed to participate in this study. Due to post-operative complications and prolonged hospitalisation in five subjects, data from 25 subjects were analysed.

Figure 1 Study flowchart

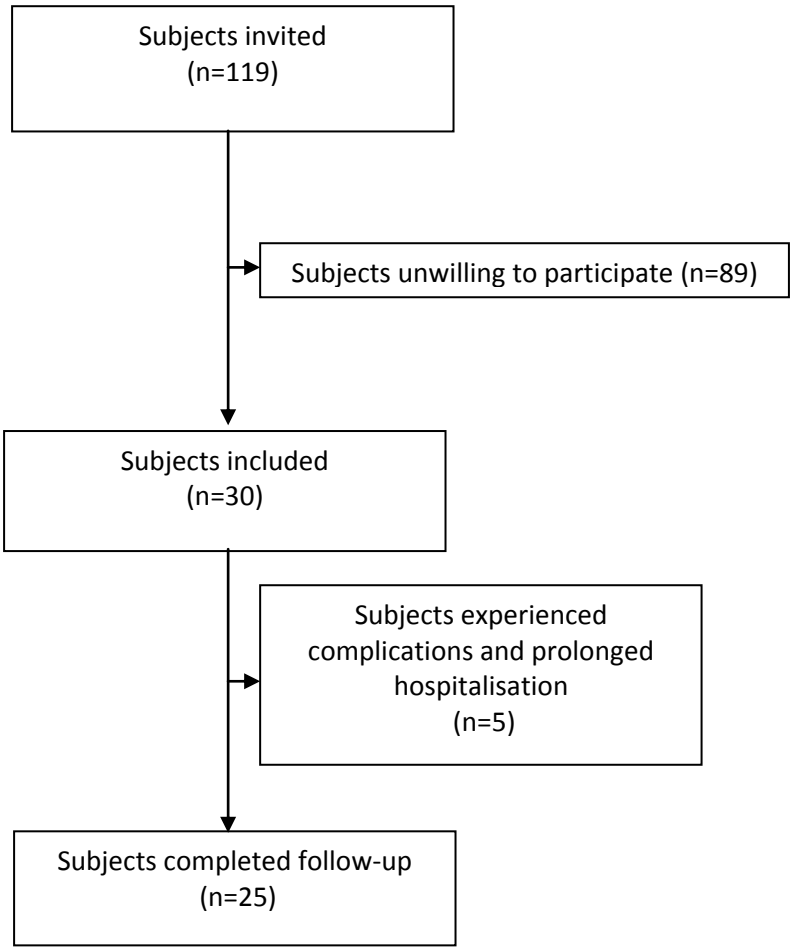
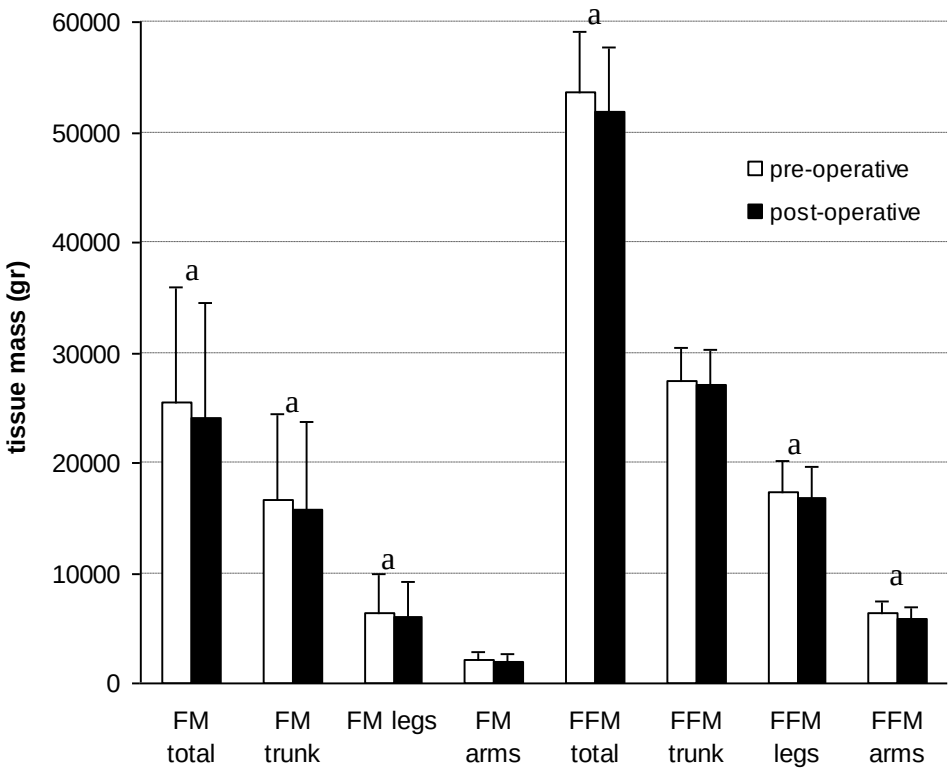


Figure 2

In Figure 2, changes in adipose tissue and fat-free mass after CABG surgery are depicted. Together with a decrease in whole-body adipose tissue mass, trunk adipose tissue mass and leg adipose tissue mass ($p<0.0025$), whole-body fat-free mass decreased by -1.9 (9.1)kg ($p<0.0025$) after CABG surgery. Significant decreases in fat-free mass were observed in the arms (by -0.5 (0.8)kg, $p<0.0025$) and legs (change of -0.5 (4.8)kg, $p<0.0025$) after CABG surgery, but not in the trunk (change of -0.7 (7.1)kg, $p>0.0025$).

Figure 2 Impact of CABG surgery on body composition



[a]Significant decrease during follow-up ($p<0.0025$).
Abbreviations: FM, fat mass, FFM, fat-free mass.

Figure 3

In Figure 3 changes in whole-body fat-free mass are depicted for each individual separately (on X-axis). A great heterogeneity in whole-body fat-free mass loss was observed. Some subjects experienced severe fat-free mass wasting (6.3kg decrease in fat-free mass) while others did not (2.7kg gain in fat-free mass).

Figure 3 Heterogeneity in whole-body fat-free tissue mass loss after CABG surgery

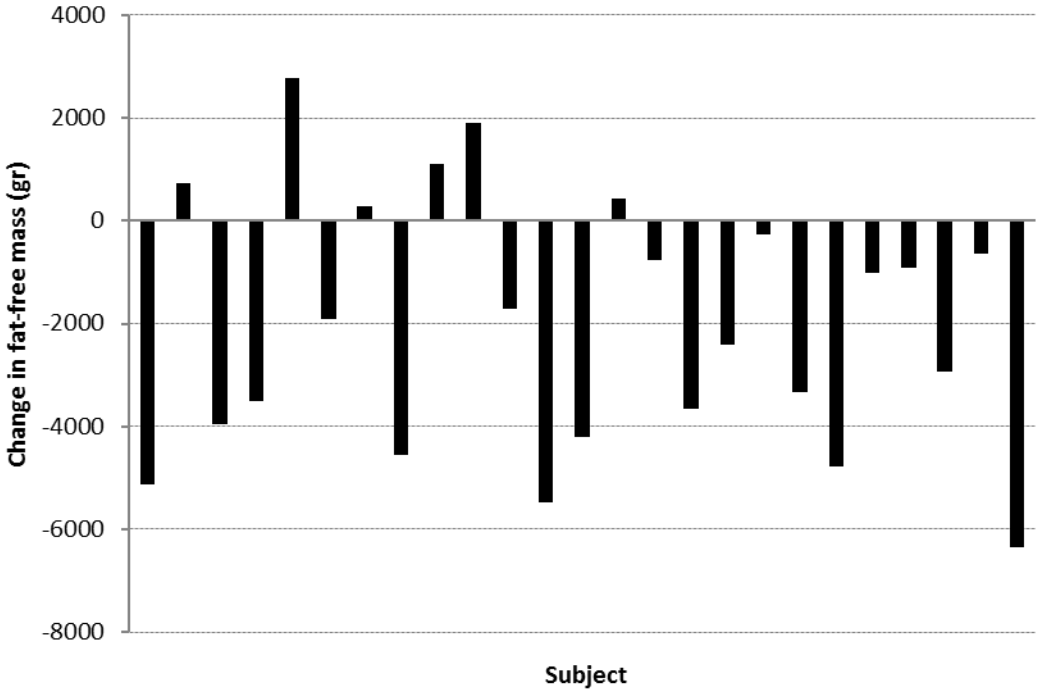


Figure 4

In Figure 4 univariate correlations between changes in fat-free mass after CABG surgery and subject characteristics/changes in blood parameters after CABG surgery are shown. Univariate correlations ($p<0.05$) were found between magnitude of whole-body fat-free mass change after surgery and time to body composition re-assessment ($r=0.43$), age ($r=0.56$), pre-operative blood c-peptide ($r=-0.53$) and cortisol ($r=0.50$) content, post-operative change in blood cortisol content ($r=-0.50$) and free androgen index ($r=0.47$).

Figure 4 Significant univariate correlations between changes in fat-free mass (in grams in X-axis) and subject characteristics/changes in blood parameters after CABG surgery (in Y-axis)