THE IMPACT OF LOW SKELETAL MUSCLE MASS IN ABDOMINAL SURGERY

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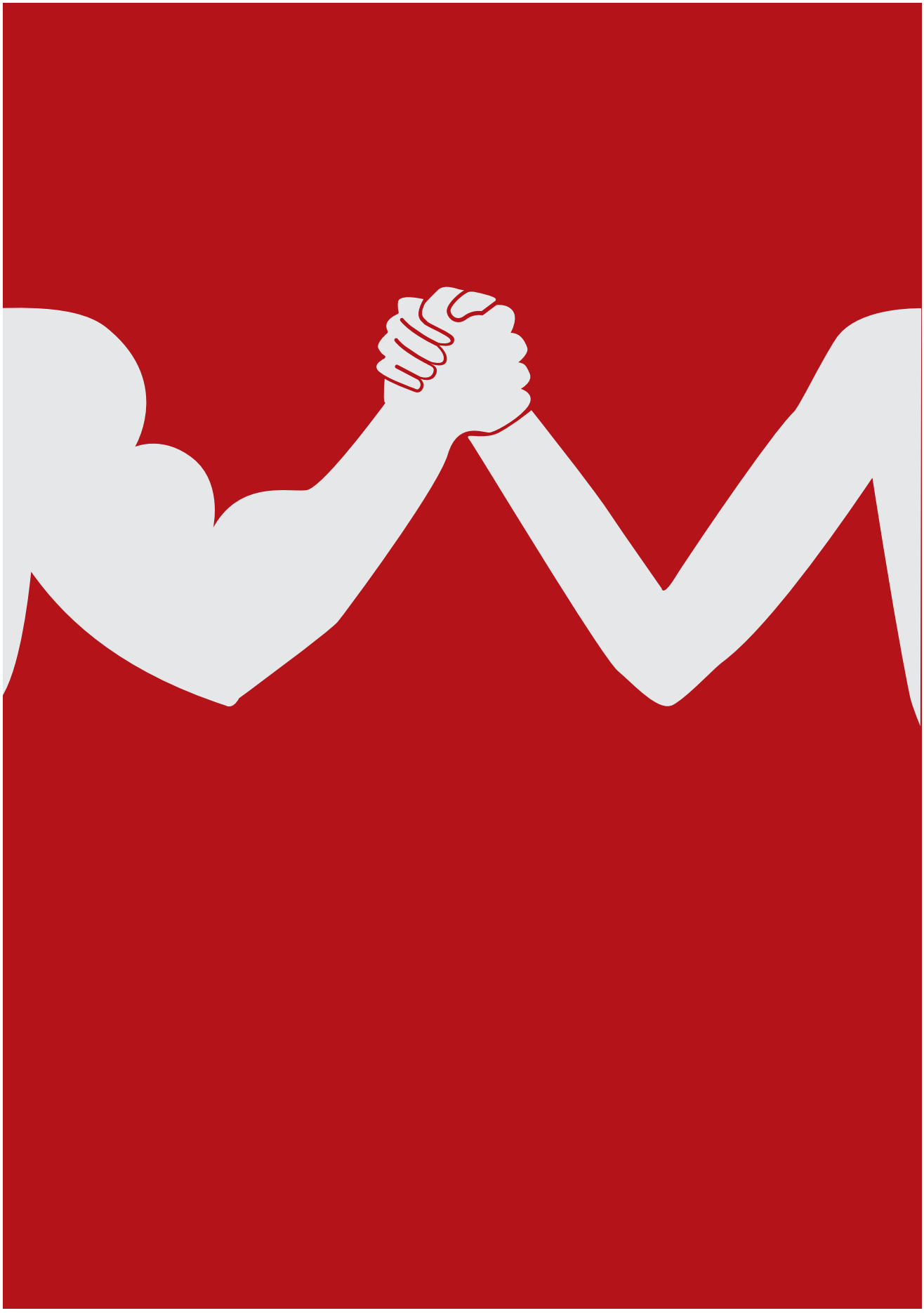

PART I

INTRODUCTION

I

CHAPTER 1

General Introduction and Aim of the Thesis

T.A. Trenning, A. Gharbharan, S. Levolger, J.L.A. van Vugt

Adapted from: The Increasing Use of Computed Tomography to Assess Body Composition and its Clinical Relevance.
Erasmus Journal of Medicine 2015 Oct; 5 (1): 51-54

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INTRODUCTION

In recent years, body composition, i.e. the relative proportions of subcutaneous adipose tissue, visceral adipose tissue, and/or skeletal muscle tissue, has extensively been studied as a predictor for outcome following oncological treatment [1], transplant surgery [2, 3], emergency and trauma surgery [4, 5], intensive care unit (ICU) admission [6], and vascular surgery [7]. Deviations in these components, which can also occur within a normal range of body weight or body mass index (BMI), are associated with individual risk and reduced survival in patients with various benign and malignant diseases [8].

The involuntary, age-dependent, depletion of skeletal muscle mass and reduction of skeletal muscle function is called sarcopenia, derived from the Greek sarx, meaning "flesh" and penia, meaning "lack of" [9]. Sarcopenia was first described in geriatric patient populations [10]. It is related to frailty, referred to as a status leading to an impaired reaction to stressors [11] and functional impairment in the elderly [12]. Skeletal muscle depletion may represent an occult condition in individuals with normal or even high body weight, as in sarcopenic obesity [13]. Moreover, skeletal muscle wasting is part of the cancer cachexia syndrome, a condition that leads to skeletal muscle wasting with or without the loss of adipose tissue. Up to 80% of patients with advanced cancer are affected by cancer-induced cachexia, and an estimated 30% of cancer-related deaths result from cancer cachexia [14-20]. Not only increasing age and cancer, but also other diseases such as liver failure, may significantly contribute to wasting of lean body and adipose tissue mass [20].

A brief history of body composition analysis using cross-sectional imaging

A reliable method that is applicable on a large scale and with high specificity, sensitivity, and reproducibility is recommended to perform skeletal muscle mass measurements. Computed tomography (CT) and magnetic resonance imaging (MRI) are known for their specificity and precision regarding body imaging and may reveal otherwise occult muscle depletion [21].

Between 1979 and 1981, Heymsfield *et al.* were the first to report the use of CT images to measure skeletal muscle and visceral adipose tissue mass [22]. In 1986, Kvist *et al.* were the first to assess whole-body adipose tissue volumes with CT, using just several slices [23]. Shen *et al.* showed a high correlation between two-dimensional abdominal skeletal muscle and adipose tissue areas measured on just a single slice and their respective three-dimensional total body volumes in a large sample of diverse subjects in 2004 [24].

In most studies that have later been performed, the third lumbar vertebra (L3) was chosen as the level to perform these measurements as a standardized landmark [1, 13]. At this level, the rectus abdominis, obliquus internus abdominis, obliquus externus abdominis, transversus abdominis, psoas major, quadratus lumborum, and the erector spinae muscles are visible and can be manually selected using specialized software. Predefined radio density ranges for muscle are used to discriminate between tissues. For muscle tissue the standard unit for radio density is used, being -30 to 150 Hounsfield units (HU) .Air is predefined as very radiolucent with a radio density of -1000 HU, pure water is 0 HU and metals +1000 HU. Since intra-abdominal organs may be within the same range of radio density as muscle, the approximate area of the muscles must be manually selected. These measurements are defined as the cross-sectional muscle area at lumbar level 3, adjusted for body height, resulting in the L3-muscle index (cm^2/m^2) [1].

In 2008, Prado *et al.* were the first to show that a low L3 muscle index was related to impaired outcome in patients suffering from malignancies of the upper respiratory and digestive tract [1]. They introduced the term sarcopenia in oncology. Although strictly speaking, low skeletal muscle mass alone does not comply with the definition of sarcopenia of the European Working Group on Sarcopenia in Older People (EWGSOG), since it also includes low skeletal muscle function or strength [25]. Nevertheless, this term is generally used in surgical and oncological literature to describe low skeletal muscle mass.

Advantages and limitations of CT images to measure skeletal muscle mass

The CT-based method provides more detail than the formerly used dual X-ray absorptiometry (DXA) and bioelectrical impedance analysis (BIA) [26]. A great advantage of CT compared with these modalities is that CT is routinely performed in many oncological and surgical patients as part of the diagnostic work-up. As the total body is rarely depicted in this setting, the cross sectional area at the L3 level, which correlates well with the total muscle mass proves its use. Moreover, skeletal muscle radio density can be measured [24, 27]. Low radio density represents a high amount of intramuscular adipose tissue [21].

Despite the fact that CT images are specific and precise and are considered the gold standard for body composition measurements by many authors [13, 28, 29], it is not recommended as a standard method to determine body composition in all populations. Besides extra expenses and logistics, patients are exposed to additional radiation. However, as CT is routinely used in practice for patients with abdominal cancers, this method does not add costs and does not entail additional patient radiation exposure. Thus, it is currently recommended as the method of choice for research purposes [20].

AIM OF THIS THESIS

Although perioperative outcome has greatly improved during the last decades by the introduction of new surgical techniques and enhanced recovery after surgery (ERAS) programs [30, 31], preoperative risk assessment remains of utmost importance to further improve outcomes and adapt patient-tailored treatment strategies. Considering the increasing age of the population, the increasing incidence of cancer, and the increased surgical and medical treatment options, skeletal muscle mass could be an important addition used for risk assessment or as a therapeutic target to improve treatment outcomes. Therefore, the aim of this thesis was to investigate the applicability of skeletal muscle mass measurements and to define the relevance of decreased skeletal muscle mass in surgical oncology and liver transplant patients.

REFERENCES

1. Prado CM, Lieffers JR, McCargar LJ, Reiman T, Sawyer MB, Martin L, et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. Lancet Oncol 2008; 9: 629-35.
2. Englesbe MJ, Patel SP, He K, Lynch RJ, Schaubel DE, Harbaugh C, et al. Sarcopenia and mortality after liver transplantation. Journal of the American College of Surgeons 2010; 211: 271-8.
3. Streja E, Molnar MZ, Kovesdy CP, Bunnapradist S, Jing J, Nissenson AR, et al. Associations of pretransplant weight and muscle mass with mortality in renal transplant recipients. Clin J Am Soc Nephrol 2011; 6: 1463-73.
4. Du Y, Karvellas CJ, Baracos V, Williams DC, Khadaroo RG, Acute C, et al. Sarcopenia is a predictor of outcomes in very elderly patients undergoing emergency surgery. Surgery 2014; 156: 521-7.
5. Fairchild B, Webb TP, Xiang Q, Tarima S, Brasel KJ. Sarcopenia and Frailty in Elderly Trauma Patients. World J Surg 2014.
6. Moisey LL, Mourtzakis M, Cotton BA, Premji T, Heyland DK, Wade CE, et al. Skeletal muscle predicts ventilator-free days, ICU-free days, and mortality in elderly ICU patients. Crit Care 2013; 17: R206.
7. Friedman J, Lussiez A, Sullivan J, Wang S, Englesbe M. Implications of sarcopenia in major surgery. Nutr Clin Pract 2015; 30: 175-9.
8. Yip C, Dinkel C, Mahajan A, Siddique M, Cook GJ, Goh V. Imaging body composition in cancer patients: visceral obesity, sarcopenia and sarcopenic obesity may impact on clinical outcome. Insights Imaging 2015; 6: 489-97.
9. Rolland Y, Abellan van Kan G, Gillette-Guyonnet S, Vellas B. Cachexia versus sarcopenia. Current opinion in clinical nutrition and metabolic care 2011; 14: 15-21.
10. Rosenberg IH, Roubenoff R. Stalking sarcopenia. Annals of internal medicine 1995; 123: 727-8.
11. Makary MA, Segev DL, Pronovost PJ, Syin D, Bandeen-Roche K, Patel P, et al. Frailty as a predictor of surgical outcomes in older patients. Journal of the American College of Surgeons 2010; 210: 901-8.
12. Morley JE, Baumgartner RN, Roubenoff R, Mayer J, Nair KS. Sarcopenia. J Lab Clin Med 2001; 137: 231-43.
13. Prado CM, Birdsell LA, Baracos VE. The emerging role of computerized tomography in assessing cancer cachexia. Curr Opin Support Palliat Care 2009; 3: 269-75.
14. Acharyya S, Butchbach ME, Sahenk Z, Wang H, Saji M, Carathers M, et al. Dystrophin glycoprotein complex dysfunction: a regulatory link between muscular dystrophy and cancer cachexia. Cancer cell 2005; 8: 421-32.

15. Fearon KC. Cancer cachexia: developing multimodal therapy for a multidimensional problem. Eur J Cancer 2008; 44: 1124-32.

16. Loberg RD, Bradley DA, Tomlins SA, Chinnaiyan AM, Pienta KJ. The lethal phenotype of cancer: the molecular basis of death due to malignancy. CA: a cancer journal for clinicians 2007; 57: 225-41.

17. Tisdale MJ. Mechanisms of cancer cachexia. Physiological reviews 2009; 89: 381-410.

18. Evans WJ. Skeletal muscle loss: cachexia, sarcopenia, and inactivity. The American journal of clinical nutrition 2010; 91: 1123S-7S.

19. Evans WJ, Morley JE, Argiles J, Bales C, Baracos V, Guttridge D, et al. Cachexia: a new definition. Clin Nutr 2008; 27: 793-9.

20. Fearon K, Strasser F, Anker SD, Bosaeus I, Bruera E, Fainsinger RL, et al. Definition and classification of cancer cachexia: an international consensus. Lancet Oncol 2011; 12: 489-95.

21. Martin L, Birdsell L, Macdonald N, Reiman T, Clandinin MT, McCargar LJ, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol 2013; 31: 1539-47.

22. Heymsfield SB, Wang Z, Baumgartner RN, Ross R. Human body composition: advances in models and methods. Annu Rev Nutr 1997; 17: 527-58.

23. Kvist H, Sjostrom L, Tylen U. Adipose tissue volume determinations in women by computed tomography: technical considerations. Int J Obes 1986; 10: 53-67.

24. Shen W, Punyanitya M, Wang Z, Gallagher D, St-Onge MP, Albu J, et al. Total body skeletal muscle and adipose tissue volumes: estimation from a single abdominal cross-sectional image. J Appl Physiol (1985) 2004; 97: 2333-8.

25. Cruz-Jentoft AJ, Baeyens JP, Bauer JM, Boirie Y, Cederholm T, Landi F, et al. Sarcopenia: European consensus on definition and diagnosis: Report of the European Working Group on Sarcopenia in Older People. Age Ageing 2010; 39: 412-23.

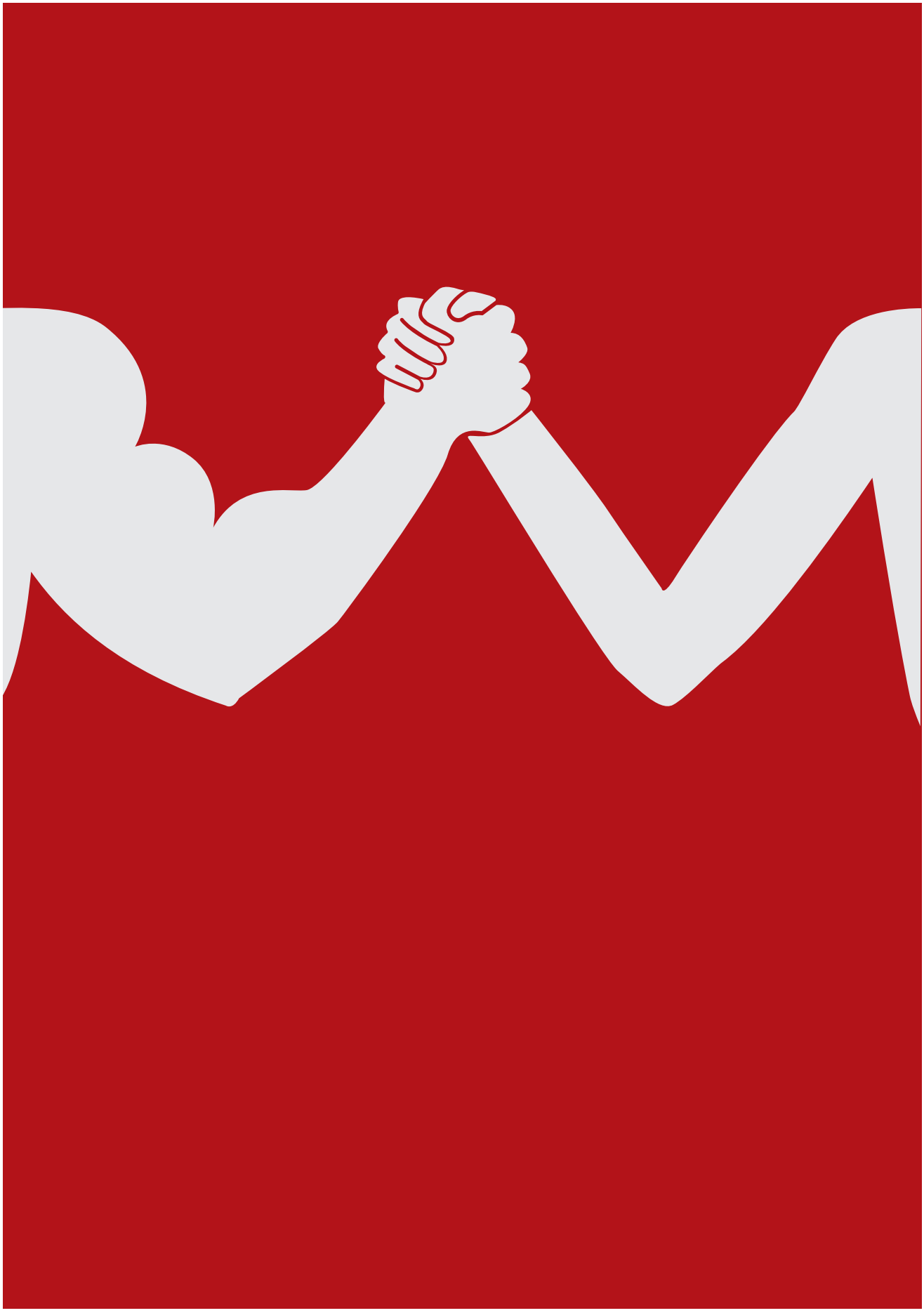
26. Mourtzakis M, Prado CM, Lieffers JR, Reiman T, McCargar LJ, Baracos VE. A practical and precise approach to quantification of body composition in cancer patients using computed tomography images acquired during routine care. Appl Physiol Nutr Metab 2008; 33: 997-1006.

27. Shen W, Punyanitya M, Wang Z, Gallagher D, St-Onge MP, Albu J, et al. Visceral adipose tissue: relations between single-slice areas and total volume. The American journal of clinical nutrition 2004; 80: 271-8.

28. Giusto M, Lattanzi B, Albanese C, Galtieri A, Farcomeni A, Giannelli V, et al. Sarcopenia in liver cirrhosis: the role of computed tomography scan for the assessment of muscle mass compared with dual-energy X-ray absorptiometry and anthropometry. Eur J Gastroenterol Hepatol 2015; 27: 328-34.

29. Prado CM. Body composition in chemotherapy: the promising role of CT scans. Current opinion in clinical nutrition and metabolic care 2013; 16: 525-33.

30. Tegels JJ, De Maat MF, Hulsewe KW, Hoofwijk AG, Stoot JH. Improving the outcomes in gastric cancer surgery. World J Gastroenterol 2014; 20: 13692-704.
31. van Vugt JL, Reisinger KW, Derikx JP, Boerma D, Stoot JH. Improving the outcomes in oncological colorectal surgery. World J Gastroenterol 2014; 20: 12445-57.



PART II

SKELETAL MUSCLE MASS MEASUREMENTS USING COMPUTED TOMOGRAPHY

II

CHAPTER 2

A Comparative Study of Software Programs for Cross-Sectional Skeletal Muscle and Adipose Tissue Measurements on Abdominal Computed Tomography Scans of Rectal Cancer Patients

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ABSTRACT

Background: The association between body composition (e.g., sarcopenia or visceral obesity) and treatment outcomes, such as survival, using single-slice computed tomography (CT) based measurements has recently been studied in various patient groups. These studies have been conducted with different software programs, each with their specific characteristics, of which the inter-observer, intra-observer and inter-software correlation are unknown. Therefore, a comparative study was performed.

Methods: Fifty abdominal CT scans were randomly selected from 50 different patients and independently assessed by two observers. Cross-sectional muscle area (CSMA, i.e. rectus abdominis, oblique and transverse abdominal muscles, paraspinal muscles and the psoas muscle), visceral adipose tissue area (VAT) and subcutaneous adipose tissue area (SAT) were segmented by using standard Hounsfield unit ranges and computed for regions of interest. The inter-software, intra-observer, and inter-observer agreement for CSMA, VAT, and SAT measurements using FatSeg, OsiriX, ImageJ, and SliceOmatic were calculated using intra-class correlation coefficients (ICC) and Bland-Altman analyses. Cohen's κ was calculated for the agreement of sarcopenia and visceral obesity assessment. The Jaccard similarity coefficient was used to compare the similarity and diversity of measurements.

Results*:* Bland-Altman analyses and intra-class correlation coefficients indicated that the CSMA, VAT, and SAT measurements between the different software programs were highly comparable (ICC 0.979-1.000, $p<0.001$). All programs adequately distinguished between the presence or absence of sarcopenia ($\kappa=0.88$-0.96 for one observer and all $k=1.00$ for all comparisons of the other observer) and visceral obesity (all $\kappa=1.00$). Furthermore, excellent intra-observer (ICC 0.999-1.000, $p<0.0001$) and inter-observer agreement (ICC 0.988-0.999, $p<0.0001$) for all software programs were found. Accordingly, excellent Jaccard similarity coefficients were found for all comparisons (mean ≥0.964).

Conclusions: FatSeg, OsiriX, ImageJ, and SliceOmatic showed an excellent agreement for CSMA, VAT and SAT measurements on abdominal CT scans. Furthermore, excellent inter- and intra-observer agreement were achieved. Therefore, results of studies using these different software programs can reliably be compared.

INTRODUCTION

Biological frailty and analytic morphomics (i.e. body composition) have increasingly gained interest in recent years in relation to treatment outcomes, such as complications and (disease-free) survival [1, 2]. Frailty, a state of increased vulnerability towards stressors, leads to an increased risk of developing adverse health outcomes [3] and is an important predictor of complications after interventional procedures, such as surgery and chemotherapy [4-7]. For example, frail patients undergoing colorectal surgery have a fourfold increased risk to develop major postoperative complications [5]. One of the hallmark signs of frailty is sarcopenia, the involuntary depletion of skeletal muscle mass [8-11]. It is estimated that up to 25% of persons under 70 years of age and over 50% of persons of 80 years and older experience sarcopenia [12]. In addition, up to 80% of patients with advanced cancer are affected by cancer-induced cachexia, a clinical condition that also results in skeletal muscle wasting with or without the loss of body fat [13-15]. Patients with cachexia are more prone to a reduced therapy effect [16] and patients with low skeletal muscle mass experience increased chemotherapy toxicity [17, 18]. This ultimately results in death in nearly one third of all cancer patients [19-22].

Over the last years, numerous studies have used abdominal computed tomography (CT) scans to quantify skeletal muscle mass, for example in clinical [17, 18, 23-25] and surgical oncology [26], vascular surgery [27], and transplantation surgery [28, 29] patients. Furthermore, multiple studies measured visceral and/or subcutaneous adipose tissue on CT scans [30-33]. However, different software programs have been used to perform these body composition analyses, such as FatSeg [33], OsiriX [7], ImageJ [24], and SliceOmatic [23]. To be able to adequately compare study results, the comparability of these various software programs should be known. Therefore, the aim of this study was to investigate the agreement of these four different software packages for the assessment of cross-sectional skeletal muscle and subcutaneous and visceral adipose tissue measurements on abdominal CT scans.

METHODS

Patients

Fifty abdominal CT scans of patients who were scheduled for rectal cancer resection at Erasmus University Medical Center (Rotterdam, The Netherlands) between 2005 and 2012 were randomly selected. All CT scans were routinely performed as part of the preoperative diagnostic work up or assessment of down staging after neo-adjuvant therapy. Only one CT scan was used per patient. None of the patients had an ostomy, abdominal wall deformity, abdominal wall tumor, or a CT scan with artefacts at the level of L3 that could potentially influence measurements. Self-reported weight and height in the preoperative work-up were retrospectively collected from electronic patient files.

Skeletal muscle and adipose tissue area measurements

The cross-sectional skeletal muscle area (CSMA), subcutaneous adipose tissue area (SAT) and visceral adipose tissue area (VAT) (cm^2), including renal adipose tissue, were measured at the mid third lumbar vertebra (L3) level on a slice showing both transversal processes. CSMA measurements included the following muscles: psoas, paraspinal, transverse abdominal, external oblique, internal oblique and rectus abdominis. All abdominal CT scans were assessed on identical slices in a random order by two medically trained observers (AG [observer A] and JLAvV [observer B]), with great knowledge about radiological anatomy and extensive experience in skeletal muscle and adipose tissue area measurements using various software programs. Observer A performed measurements twice on identical *a priori* selected slices, whereas observer B performed a second reading without *a priori* selected slice numbers. The observers were blinded for each other's measurements and for patient details. For each observer the time interval between two readings in the same patient with different software programs was at least one week. This resulted in an interval of at least four weeks between two readings within one patient with the same software program. Only the first reading of observer B was used for the inter-software and inter-observer comparisons.

The CSMA was corrected for height squared (m^2), resulting in the L3 muscle index (SMI, cm^2/m^2). Patients were classified as having sarcopenia or not having sarcopenia according to previously described cut-off values (52.4 cm^2/m^2 for men and 38.5 cm^2/m^2 for women) [23]. Predefined cut-off values for visceral adipose tissue area to define visceral obesity of 163.8 cm^2 for men and 80.1 cm^2 for women were used [34]. For subcutaneous adipose tissue no cut-off values have been reported in the literature.

Four software programs were compared: FatSeg (developed by the Biomedical Imaging Group Rotterdam of Erasmus MC, Rotterdam, The Netherlands, using MeVisLab (Mevis Medical Solutions, Bremen, Germany)), OsiriX (Pixmeo SARL, Geneva, Switzerland), ImageJ (National Institutes of Health, Bethesda, Maryland, USA), and SliceOmatic (TomoVision, Magog, Canada). CSMA, VAT, and SAT were segmented using standard Hounsfield Unit (HU) thresholds in all four software programs. An intensity window between -30 HU and +150 HU was used for skeletal muscle tissue [35]. For adipose tissue an intensity window between -190 HU and -30 HU was used [36]. Since the tissue of interest is manually selected, competency in anatomic radiology is a prerequisite for these measurements.

FatSeg

FatSeg is an in-house developed software program to perform soft tissue measurements on CT scans and was developed using the MeVisLab development environment for medical image processing and visualization version 2.4 (available from http://www.mevislab.de). Inner and outer contours of aforementioned skeletal muscle and adipose regions were manually traced. The skeletal muscle and adipose tissue areas were computed automatically using the preset HU intensity thresholds, and expressed in cm^2. Intraluminal contents initially marked as adipose tissue were manually erased. Cutaneous tissue was included in the SAT measurement. Measurements were performed on a 3.2 GHz Intel® Core™ i5 Dell (Dell Inc., Round Rock, TX, USA) personal computer.

OsiriX

The open-source 32-bit edition of OsiriX version 5.8.5 (available from http://www.osirix-viewer.com) was used. The "Grow Region (2D/3D Segmentation)" tool was used to semi-automatically select skeletal muscle and adipose tissue regions within our preset HU intensity thresholds. Non-skeletal muscle tissue regions adjacent to skeletal muscle were manually removed from the area selection using the brush option. The brush option was also used to manually erase intraluminal areas with contents having radiological density between -190 and -30 HU, resembling fatty content. Cutaneous tissue was not included in the SAT measurement. The skeletal muscle and adipose tissue areas were computed automatically and expressed in cm^2 using a 1.3 GHz Intel® Core™ i5 MacBook Air (Apple Inc., Cupertino, CA, USA) and computer mouse.

ImageJ

ImageJ version 1.48 is a freely downloadable public domain software program developed by the National Institutes of Health for image processing and analyzing (available from http://rsbweb.nih.gov/ij/download.html). First, manual delineation of the outer contour

of the abdominal wall and paraspinal muscles was performed and the surface area of tissue with an attenuation between -30 and +150 HU was computed automatically (mm^2) and manually divided by 100, resulting in cm^2. Second, delineation of the inner contour of the abdominal wall, paraspinal and psoas muscles was performed in a similar fashion to allow for subsequent correction of intra-abdominal content with attenuation between the preset HU intensity thresholds. The inner contour was manually subtracted from the outer contour surface area, resulting in the cross-sectional skeletal muscle area (cm^2) [37]. The subcutaneous adipose tissue area measurements were performed in a similar manner as the muscle measurements, whereas visceral adipose tissue area measurements were performed by delineating a contour through the inner contour of the abdominal wall muscles, psoas muscles and vertebrae followed by manual erasing of intraluminal fatty content. Cutaneous tissue was included in the SAT measurement. A 3.2 GHz Intel® Core™ i5 Dell (Dell Inc., Round Rock, TX, USA) personal computer was used.

SliceOmatic

SliceOmatic (TomoVIsion, Magog, Canada) version 5.0 (64 bit; available from http://www.tomovision.com/) was used. Tissue was semi-automatically selected with the 'Region Growing' mode using the 'Grow 2D' and 'Paint' tools. Non-skeletal muscle tissue regions adjacent to skeletal muscle having radiological density between the predefined HU thresholds were manually erased using the 'Paint' tool. Cutaneous tissue was included in the SAT measurement. A 3.2 GHz Intel® Core™ i5 Dell (Dell Inc., Round Rock, TX, USA) personal computer was used.

Cutaneous tissue disclosure

In OsiriX, cutaneous tissue is not included in the SAT measurement, because this is not automatically selected using the ''Grow Region (2D/3D Segmentation)'' tool. SliceOmatic also allows to exclude encompassed skin. However, not all software programs allow to reliably exclude cutaneous tissue from SAT as a consequence of their measurement method: the delineation of tissue of interest using inner and outer contours. Consequently, to ensure highly comparable measurements in three rather than two software programs, cutaneous tissue was included in the SAT measurements with SliceOmatic. A comparison of SAT measurements using SliceOmatic with and without the inclusion of cutaneous tissue resulted in a median difference of 2.3% (interquartile range 0.8-3.8) and was considered acceptable.

Statistical analysis

Continuous data are presented as mean with the standard error of the measurement (SEM). Normality was tested using the Shapiro-Wilk test. Differences between the different software packages and within and between observers were compared using the paired samples t-test for normally distributed data and the Wilcoxon signed rank test for data that was not normally distributed. The inter-software and inter- and intra-observer agreement for the cross-sectional skeletal muscle, visceral adipose tissue and subcutaneous adipose tissue measurements were calculated using intra-class correlation coefficients (ICC) with 95% confidence intervals (CI) using a two-way mixed single measures model with absolute agreement. For the inter-observer correlation, the reading of observer B was compared with reading 1 of observer A. Ninety-five % limits of agreement were determined to investigate the agreement between the various software programs, according to the method described by Bland and Altman [38]. The presence of proportional systematic bias was determined by linear regression analysis of the difference and mean of two measurements. The inter-software and inter- and intra-observer agreement of the assessment of sarcopenia and visceral obesity were analyzed using Cohen's κ coefficients. The ICC and Cohen's κ coefficients were interpreted as poor (0.00-0.49), fair to good (0.50-0.74) and excellent (0.75-1.00), as proposed by Shrout and Fleiss [39]. The Jaccard similarity coefficient, ranging from 0-1, was used to compare the similarity and diversity of measurements by dividing the area of the intersection by the size of the union of two measurements [40]. An overlay of two measurements was created and the Jaccard similarity coefficient was calculated using MeVisLab version 2.7.1 (MeVis Medical Solutions AG, Bremen, Germany). A Jaccard similarity coefficient of 1 represents perfect overlap of two samples, whereas 0 represents no overlap. Two-tailed p-values <0.05 were considered statistically significant. All statistical analyses were performed using IBM SPSS Statistics for Windows version 21.0 (IBM Corp. Armonk, NY, USA).

RESULTS

Patients

The study population consisted of 29 males (58%) and 21 females (42%) with a median age of 62 years (range 33 - 81) and a median body mass index (BMI) of 24.6 kg/m^2 (range 16.5 - 38.8). Ten patients had stage II (20.0%), 24 stage III (48.0%) and 15 stage IV (30.0%) rectal cancer. Tumor stage was unknown for one patient. The mean CSMA, VAT, and SAT for all measurements are provided in table 1 and table 2.

Inter-software agreement

The inter-software ICCs were excellent (≥0.999) for the CSMA, VAT, and SAT for all software programs with p-values <0.001 (table 3). Figure 1 and supplementary figures 1 and 2 show the Bland-Altman 95% limits of agreement plots, with the mean difference and 95% limits of agreement for the CSMA, VAT, and SAT for both observers. All plots show a good agreement between the various software programs. Small limits of agreement are observed in the CSMA measurements, whereas these limits of agreement are greater for the VAT and SAT measurements. Proportional systematic bias was observed between FatSeg and OsiriX for CSMA (p=0.049) for observer B (figure 1a) and between FatSeg and SliceOmatic for SAT (p=0.031) for observer A (supplementary figure 2c). Furthermore, proportional systematic bias was frequently observed between programs for VAT measurements (supplementary figure 1). Comparable results were achieved when non *a priori* selected slices of observer B were analyzed (data not shown). The mean Jaccard similarity coefficients for the inter-software comparisons are summarized in table 4 and depicted in figure 2.

Intra-observer and inter-observer agreement

The ICCs for the intra-observer agreement of observer A were all 0.979 or higher for the different software programs, approaching perfect correlation (table 1). The ICCs for the inter-observer agreement also approached perfect agreement (all ≥0.999, see table 2). The mean CSMA was significantly lower for observer A compared with observer B for all software programs. A significantly higher mean VAT of observer A was found using FatSeg (149.9 cm^2 versus 148.7 cm^2, p<0.001) and ImageJ (148.6 cm^2 versus 148.4 cm^2, p=0.015) compared with observer B, whereas the mean VAT of both observers did not significantly differ for OsiriX (p=0.133) and SliceOmatic (p=0.412). The mean SAT did significantly differ for FatSeg (158.9 cm^2 versus 159.2 cm^2, p=0.005) between the observers. Comparable results were observed when non *a priori* selected slices of reading 2 of observer B were used for analyses (data not shown). The mean Jaccard

similarity coefficients for the inter- and intra-observer comparisons are summarized in table 5 and depicted in supplementary figures 3 and 4. All remaining worst Jaccard similarity coefficients are provided in supplementary figure 5.

Table 1. Mean cross-sectional skeletal muscle and visceral and subcutaneous adipose tissue area (cm^2) measurements and intra-observer agreement indices (i.e. ICC) using FatSeg, OsiriX, ImageJ, and SliceOmatic of observer A. SEM, standard error of measurement; ICC, inter- and intra-class correlation coefficients; CI confidence intervals.

Software	Observer A						
	Reading 1 (cm^2)	SEM	Reading 2 (cm^2)	SEM	Mean difference (95% CI)	p-value	ICC (95% CI)
Skeletal muscle area							
FatSeg	139.0	5.2	139.3	5.2	-0.3 (-0.6; 0.0)	0.072*	0.999 (0.999-1.000)
OsiriX	139.4	5.2	138.7	5.1	0.7 (0.4; 1.0)	<0.001*	0.999 (0.999-1.000)
ImageJ	139.0	5.2	139.3	5.1	-0.3 (-0.6; -0.1)	0.013*	1.000 (0.999-1.000)
SliceOmatic	138.7	5.2	138.6	5.2	0.1 (-0.2; 0.4)	0.441*	1.000 (0.999-1.000)
Visceral adipose tissue area							
FatSeg	149.9	13.1	149.2	13.1	0.7 (0.3; 1.0)	<0.001#	1.000 (1.000-1.000)
OsiriX	147.6	13.0	147.3	13.0	0.3 (-0.3; 0.8)	0.220#	1.000 (1.000-1.000)
ImageJ	148.6	13.0	150.8	12.8	-2.2 (-7.5; 3.1)	0.003#	0.979 (0.964-0.988)
SliceOmatic	147.1	13.0	146.6	13.0	0.5 (0.2; 0.9)	0.004#	1.000 (1.000-1.000)
Subcutaneous adipose tissue area							
FatSeg	158.9	11.2	158.9	11.2	0.1 (-0.2; 0.3)	0.359#	1.000 (1.000-1.000)
OsiriX	155.9	11.2	155.7	11.3	0.2 (-0.1; 0.4)	0.137#	1.000 (1.000-1.000)
ImageJ	158.9	11.2	159.1	11.3	-0.2 (-0.5; 0.0)	0.201#	1.000 (1.000-1.000)
SliceOmatic	158.8	11.3	158.8	11.3	0.0 (-0.3; 0.2)	0.448#	1.000 (1.000-1.000)

Calculated with *paired-samples t-test and #Wilcoxon signed rank test

Table 2. Mean cross-sectional skeletal muscle and visceral and subcutaneous adipose tissue area (cm^2) measurements and inter-observer agreement indices (i.e. ICC) using FatSeg, OsiriX, ImageJ, and SliceOmatic of reading 1 of observer A and observer B. SEM, standard error of measurement; ICC, inter- and intra-class correlation coefficients; CI confidence intervals.

Software	Observer A		Observer B		Mean difference (95% CI)	p-value	ICC (95% CI)
	Reading 1 (cm^2)	SEM	Reading 1 (cm^2)	SEM			
Skeletal muscle area (CSMA)							
FatSeg	139.0	5.2	140.1	5.2	-1.1 (-1.4; -0.8)	<0.001*	0.999 (0.989-1.000)
OsiriX	139.4	5.2	139.7	5.1	-0.3 (-0.5; 0.0)	0.047*	1.000 (0.999-1.000)
ImageJ	139.0	5.2	139.8	5.2	-0.8 (-1.0; -0.5)	<0.001*	0.999 (0.997-1.000)
SliceOmatic	138.7	5.2	139.3	5.2	-0.6 (-0.9; -0.2)	0.006*	0.999 (0.998-1.000)
Visceral adipose tissue area (VAT)							
FatSeg	149.9	13.1	148.7	13.1	1.2 (0.8; 1.5)	<0.001#	1.000 (0.999-1.000)
OsiriX	147.6	13.0	147.3	13.0	0.3 (-0.3; 0.8)	0.133#	1.000 (1.000-1.000)
ImageJ	148.6	13.0	148.4	13.1	0.3 (-0.1; 0.6)	0.015#	1.000 (1.000-1.000)
SliceOmatic	147.1	13.0	146.9	13.0	0.2 (-0.1; 0.5)	0.412#	1.000 (1.000-1.000)
Subcutaneous adipose tissue area (SAT)							
FatSeg	158.9	11.2	159.2	11.3	-0.3 (-0.5; -0.1)	0.005#	1.000 (1.000-1.000)
OsiriX	155.9	11.2	155.8	11.3	0.1 (-0.3; 0.5)	0.918#	1.000 (1.000-1.000)
ImageJ	158.9	11.2	158.7	11.2	0.2 (-0.2; 0.5)	0.306#	1.000 (1.000-1.000)
SliceOmatic	158.8	11.3	158.5	11.2	0.2 (0.0; 0.5)	0.183#	1.000 (1.000-1.000)

Calculated with *paired-samples t-test and #Wilcoxon signed rank test

Table 3. Mean cross-sectional skeletal muscle and visceral and subcutaneous adipose tissue area (cm^2) measurements and inter-software agreement indices (i.e. ICC) using FatSeg, OsiriX, ImageJ, and SliceOmatic of reading 1 of observer B. The results of observer A are comparable with those of observer B. SEM, standard error of measurement; ICC, inter- and intra-class correlation coefficients; CI confidence intervals.

Software program	Mean difference (cm^2) (95% CI)	SEM	p-value	ICC (95% CI)
Skeletal muscle area (CSMA)				
FatSeg – OsiriX	-0.4 (-0.8; 0.0)	0.184	0.047	0.999 (0.999-1.000)
FatSeg – ImageJ	0.0 (-0.3; 0.3)	0.151	0.992	1.000 (0.999-1.000)
FatSeg – SliceOmatic	0.3 (-0.2; 0.8)	0.230	0.207	0.999 (0.998-0.999)
OsiriX – ImageJ	0.4 (0.1; 0.7)	0.161	0.023	0.999 (0.999-1.000)
OsiriX – SliceOmatic	0.7 (0.3; 1.1)	0.189	0.001	0.999 (0.998-1.000)
ImageJ – SliceOmatic	0.3 (-0.1; 0.7)	0.208	0.165	0.999 (0.999-1.000)
Visceral adipose tissue area (VAT)				
FatSeg – OsiriX	2.3 (1.6; 2.9)	0.326	<0.001	0.999 (0.995-1.000)
FatSeg – ImageJ	1.2 (0.8; 1.7)	0.203	<0.001	1.000 (0.999-1.000)
FatSeg – SliceOmatic	2.8 (2.3; 3.2)	0.238	<0.001	0.999 (0.971-1.000)
OsiriX – ImageJ	-1.0 (-1.5; -0.6)	0.237	<0.001	1.000 (0.999-1.000)
OsiriX – SliceOmatic	0.5 (0.0; 0.9)	0.229	0.044	1.000 (1.000-1.000)
ImageJ – SliceOmatic	1.5 (1.2; 1.8)	0.158	<0.001	1.000 (0.995-1.000)
Subcutaneous adipose tissue area (SAT)				
FatSeg – OsiriX	3.0 (2.5; 3.6)	0.256	<0.001	0.999 (0.948-1.000)
FatSeg – ImageJ	0.1 (-0.3; 0.4)	0.180	0.698	1.000 (1.000-1.000)
FatSeg – SliceOmatic	0.2 (-0.1; 0.5)	0.141	0.240	1.000 (1.000-1.000)
OsiriX – ImageJ	-3.0 (-3.5; -2.5)	0.260	<0.001	0.999 (0.956-1.000)
OsiriX – SliceOmatic	-2.9 (-3.3; -2.5)	0.211	<0.001	0.999 (0.932-1.000)
ImageJ – SliceOmatic	0.1 (-0.2; 0.4)	0.139	0.485	1.000 (1.000-1.000)

Table 4. Mean Jaccard indices for inter-software comparisons of reading 1 of observer A and reading 1 of observer B. CSMA, cross-sectional muscle area; VAT, visceral adipose tissue area, SAT, subcutaneous adipose tissue area.

	Mean Jaccard similarity coefficients					
	Observer A (reading 1) (range)			**Observer B (reading 1) (range)**		
	CSMA	**VAT**	**SAT**	**CSMA**	**VAT**	**SAT**
FatSeg – OsiriX	0.978 (0.940-0.997)	0.964 (0.825-0.996)	0.965 (0.928-0.976)	0.983 (0.948-0.997)	0.973 (0.886-0.997)	0.965 (0.926-0.998)
FatSeg – ImageJ	0.982 (0.935-0.996)	0.981 (0.912-0.999)	0.988 (0.900-0.999)	0.987 (0.959-0.998)	0.981 (0.903-0.998)	0.990 (0.968-0.998)
FatSeg – SliceOmatic	0.978 (0.937-0.996)	0.970 (0.908-0.997)	0.989 (0.964-0.998)	0.981 (0.927-0.996)	0.972 (0.860-0.997)	0.987 (0.960-0.998)
OsiriX – ImageJ	0.982 (0.935-0.996)	0.968 (0.856-0.995)	0.964 (0.900-0.998)	0.983 (0.948-0.997)	0.976 (0.891-0.998)	0.966 (0.927-0.997)
OsiriX – SliceOmatic	0.979 (0.941-0.997)	0.974 (0.876-0.997)	0.988 (0.900-0.998)	0.985 (0.944-0.997)	0.973 (0.884-0.998)	0.967 (0.923-0.998)
ImageJ – SliceOmatic	0.979 (0.950-0.994)	0.967 (0.809-0.995)	0.966 (0.928-0.997)	0.983 (0.932-0.997)	0.975 (0.855-0.998)	0.988 (0.965-0.999)

Table 5. Mean Jaccard indices for inter-observer (reading 1 of observer A versus reading 1 of observer B) and intra-observer comparisons (reading 1 versus reading 2 of observer A). CSMA, cross-sectional muscle area; VAT, visceral adipose tissue area, SAT, subcutaneous adipose tissue area.

	Mean Jaccard similarity coefficients					
	Inter-observer (range)			**Intra-observer (range)**		
	CSMA	**VAT**	**SAT**	**CSMA**	**VAT**	**SAT**
FatSeg	0.981 (0.949-0.997)	0.976 (0.908-0.998)	0.991 (0.969-0.999)	0.982 (0.961-1.000)	0.984 (0.916-0.999)	0.991 (0.956-1.000)
OsiriX	0.985 (0.960-0.997)	0.973 (0.835-0.997)	0.989 (0.960-1.000)	0.984 (0.953-0.997)	0.975 (0.838-0.998)	0.990 (0.967-1.000)
ImageJ	0.982 (0.931-0.993)	0.980 (0.905-0.997)	0.988 (0.899-0.999)	0.985 (0.948-1.000)	0.982 (0.891-0.998)	0.990 (0.900-1.000)
SliceOmatic	0.981 (0.939-0.997)	0.976 (0.876-0.996)	0.989 (0.959-0.999)	0.986 (0.961-0.997)	0.980 (0.901-0.998)	0.993 (0.967-1.000)

Figure 1. Bland-Altman 95% limits of agreement plots for the agreement between the various software programs (provided on the X-axes and Y-axes) for CSMA (cm^2). The dotted lines are the mean of the difference and the 95% limits of agreement (± 2 SD) between the CSMA of reading 1 of observer A and the solid lines of reading 1 of observer B.

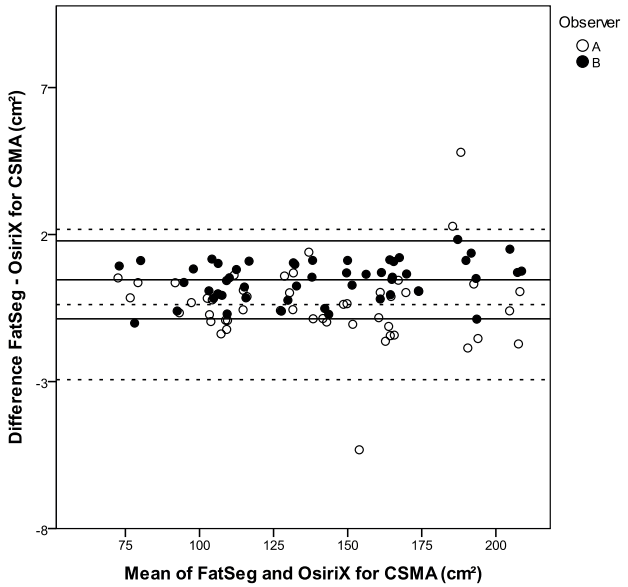

Figure 1a. There was no proportional systematic bias for observer A (p=0.908), whereas there was significant bias for observer B (p=0.049).

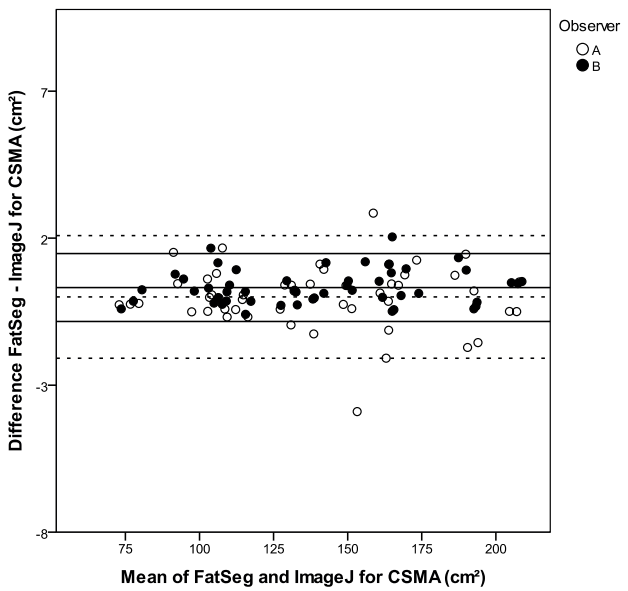

Figure 1b. There was no proportional systematic bias for any observer (p=0.738 and p=0.359).

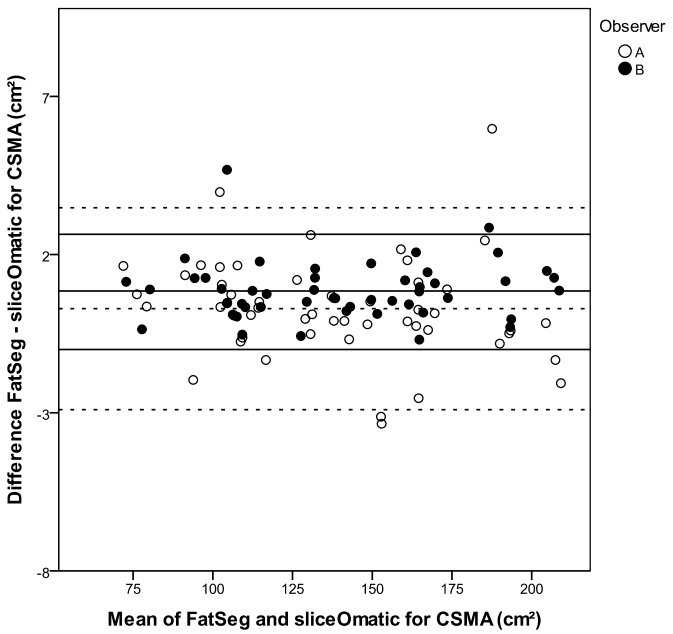

Figure 1c. There was no proportional systematic bias for any observer (p=0.238 and p=0.704).

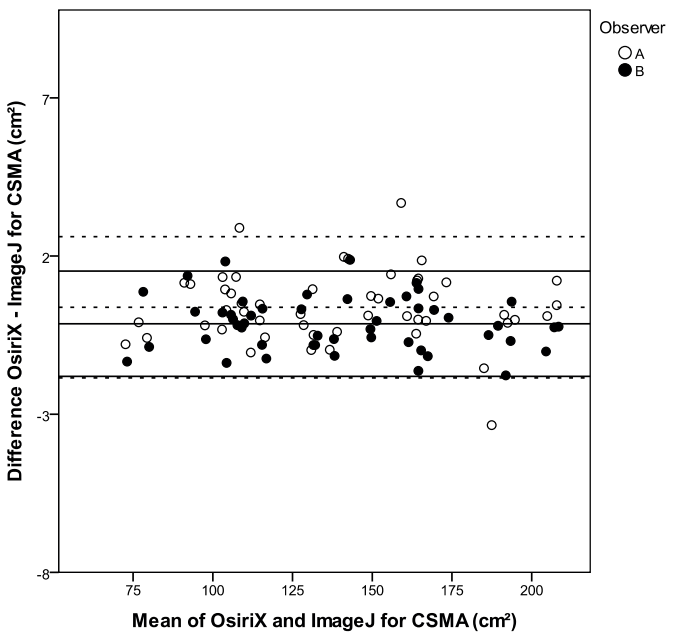

Figure 1d. There was no proportional systematic bias for any observer (p=0.857 and p=0.363).

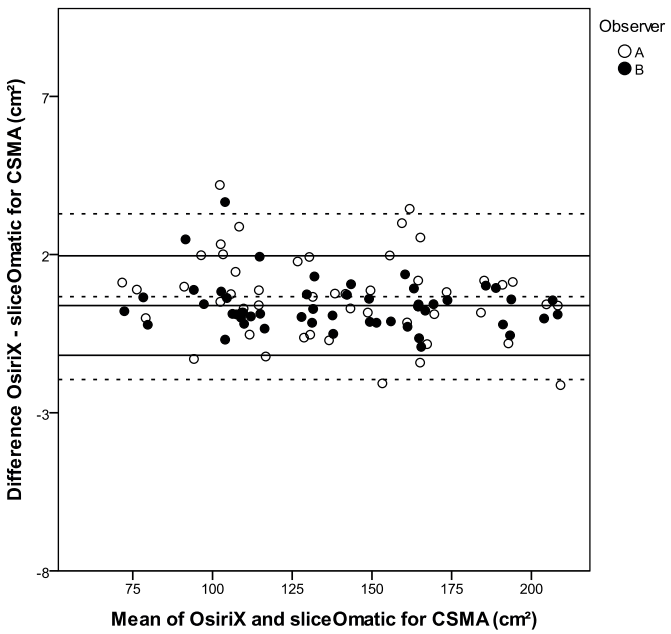

Figure 1e. There was no proportional systematic bias for any observer (p=0.185 and p=0.228).

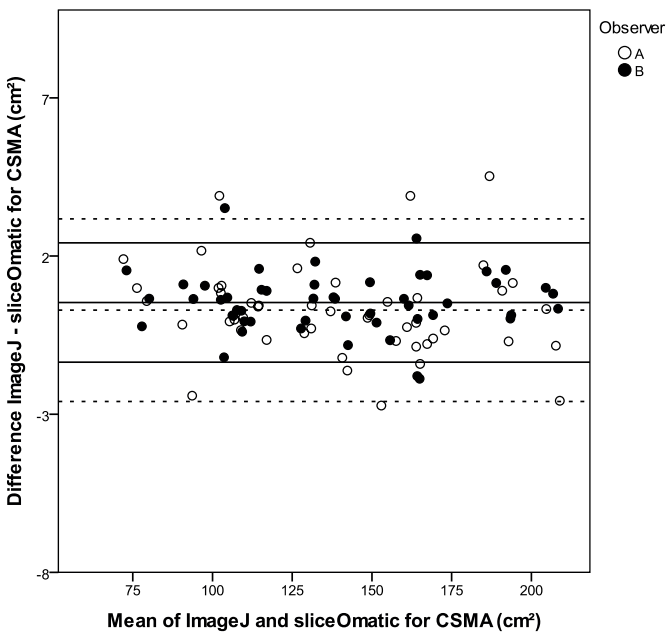

Figure 1f. There was no proportional systematic bias for any observer (p=0.289 and p=0.843).

Figure 2. Jaccard similarity coefficients (lowest and highest are shown) for inter-software comparisons of CSMA, VAT, and SAT (cm^2) measurements (reading 1 of observer B). The green area represents similarity, whereas the red area represents discrepancy in measurements.

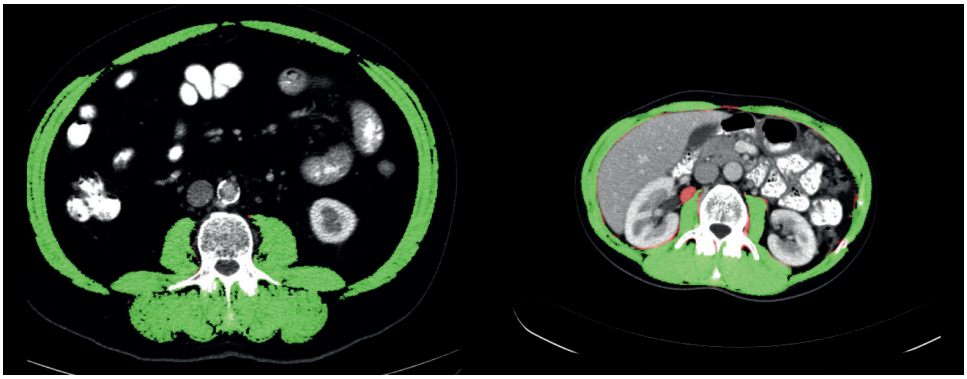

Figure 2a. The CSMA measured with FatSeg and ImageJ (1) and FatSeg and SliceOmatic (2), resulting in Jaccard similarity coefficients of 0.998 and 0.927, respectively.

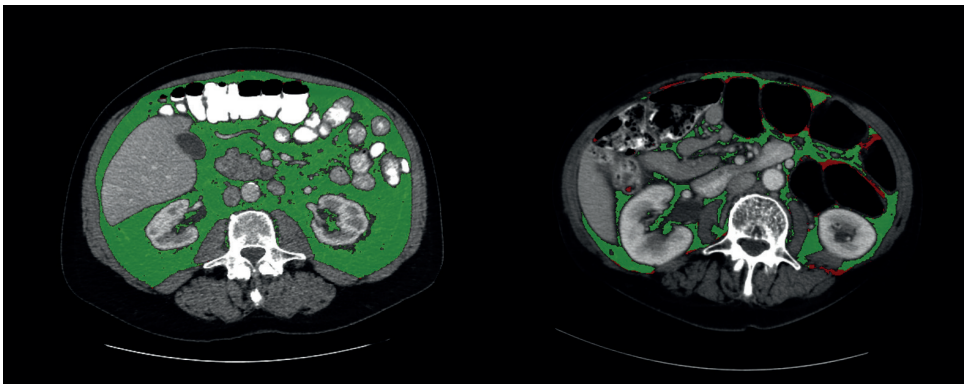

Figure 2b. The VAT measured with ImageJ and SliceOmatic (1 and 2), resulting in Jaccard similarity coefficients of 0.998 and 0.855, respectively.

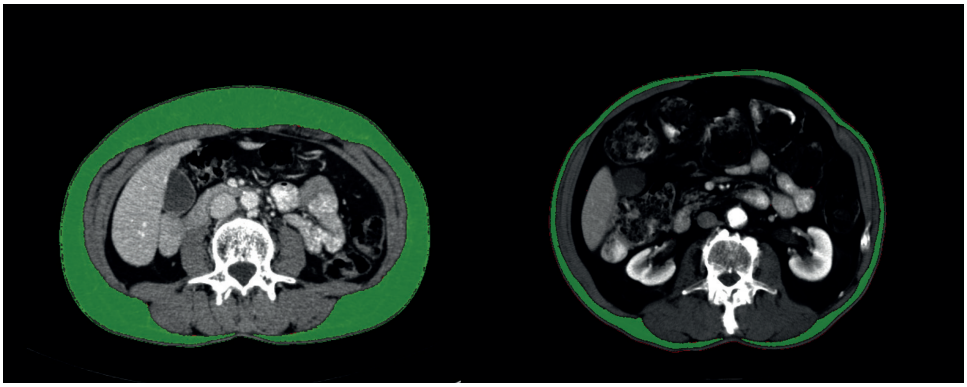

Figure 2c. The SAT measured with ImageJ and SliceOmatic (1) and OsiriX and SliceOmatic (2), resulting in Jaccard similarity coefficients of 0.999 and 0.923, respectively.

The classification of sarcopenia and visceral obesity

The inter-software Cohen's κ's of the first reading of observer A for the classification of sarcopenia were 0.96 (between FatSeg and Osirix, OsiriX and ImageJ, and ImageJ and SliceOmatic), 0.92 (between FatSeg and ImageJ, and Osirix and SliceOmatic), and 0.88 (between FatSeg and SliceOmatic). No inter-software differences were found in the classification of patients with and without sarcopenia for observer B. According to the cut-off values used, all software programs diagnosed sarcopenia in 16 men (55.2%) and 8 women (38.1%). This resulted in a Cohen's κ of 1.00 for all comparisons between software programs ($p<0.001$).

The Cohen's κ for the intra-observer agreement of sarcopenia assessment of observer A was 0.96 using FatSeg and ImageJ and 1.00 for OsiriX and SliceOmatic (all $p<0.001$).

The Cohen's κ for the inter-observer agreement (reading 1 of observer A versus observer B) of sarcopenia assessment was 0.92 for SliceOmatic, 0.96 for FatSeg and ImageJ, and 1.00 for Osirix (all $p<0.001$).

The classification of visceral obesity

In total, 17 men (58.6%) and 9 women (42.9%) were classified as visceral obese using FatSeg, OsiriX, ImageJ, and SliceOmatic in all readings. This resulted in a Cohen's κ of 1.00 for all comparisons (all $p<0.001$).

DISCUSSION

This study shows that the inter-software agreement was excellent for all software programs. Furthermore, the inter-observer and intra-observer agreements were excellent for four distinct software programs to assess CSMA, VAT, and SAT on abdominal CT scans with high Jaccard similarity coefficients.

Body composition analyses using abdominal CT scans are increasingly being performed. In multiple surgical populations, such as vascular [27], gastrointestinal [7, 33], urological [41-43], gynecological [44] and transplantation surgery [29], the association between low skeletal muscle mass and an increased risk of postoperative complications, recurrent disease, or impaired survival has been shown. Low skeletal muscle mass is also related to discharge destination in elderly trauma patients [45], associated with an increased risk of dose-limiting chemotherapy toxicity [17, 24, 25, 46] and with morbidity and mortality in various oncologic populations, such as lung cancer and melanoma patients [47, 48]. Furthermore, CT-assessed visceral obesity is associated with worse short- and long-term outcome in distinct patient populations undergoing surgery [30]. Various software programs have been used to measure body composition in these studies. The current study shows that the results of these studies can reliably be compared. Based on our findings it is likely that this is also true for other software programs which similarly compute skeletal muscle area by quantifying selected voxels within preset HU intensity thresholds (e.g., studies that used software programs designed in MATLAB [MathWorks, Natick, MA, USA] [49]).

Software programs for various body composition measurements on CT images, such as adipose tissue surface area, skeletal muscle tissue surface area and liver volumetric measurements, have been compared in multiple previous studies, demonstrating high levels of agreement [50-53]. Excellent agreement levels between SliceOmatic and ImageJ [54], as well as between observers using SliceOmatic [55] for CSMA measurements have previously been reported. Furthermore, excellent agreement levels between OsiriX and ImageJ have been observed for paraspinal muscle measurements on magnetic resonance images [56]. Nevertheless, this is the first study to compare multiple software programs for the measurement of CSMA, VAT, and SAT, showing that previous studies investigating the association between skeletal muscle mass on the one side and visceral or subcutaneous adipose tissue on the other side, and patient outcomes can reliably be compared.

The skeletal muscle area (cm^2) measured at a single cross-sectional CT image at the level of the third lumbar vertebra (L3) is linearly related to total body skeletal muscle mass [57] and is therefore corrected for height squared (m^2), as is conventional for

body composition measures. This results in the L3 muscle index (cm^2/m^2) [18]. Another frequently used method is measuring the total psoas area (TPA) [27]. The principle of TPA measurements is identical to L3 muscle area measurements, using single cross-sectional CT images. Therefore, the findings of this study may be extrapolated to TPA measurements as well. Nevertheless, this should be confirmed in a future study.

Significant differences were observed between the mean skeletal muscle areas within and between observers. However, these mean differences are small and consequently not clinically relevant. Differences in individual measurements resulted, for instance, from the incorrect annotation of skeletal muscle tissue (see figure 2a2 for an example of an intra-observer difference). However, we decided not to correct measurements in retrospect to show inter- and intra-observer agreements. In our opinion, this study reflects daily practice, with observers who have excellent (radiological) anatomical knowledge performing body composition measurements. Regardless of these human errors and some inter- and intra-observer differences, high comparability between software programs was observed.

Significant differences between VAT measurements were also observed with greater mean differences between software. This could due to the greater complexity of the measurement technique, as intraluminal content (i.e. fat in stool) needs to be manually erased. The greatest significant mean differences in SAT could partly been explained by the fact that in OsiriX the cutaneous adipose tissue in not included in the SAT, in contrast to the other software programs. Furthermore, every tissue of interest needs to be manually selected in OsiriX, in contrast to the other programs in which methods of delineating or a painting brush can be used to select regions of interest.

Significant differences in the mean VAT (FatSeg and ImageJ) within observer A and in the mean VAT (FatSeg OsiriX, and SliceOmatic) and SAT (FatSeg, OsiriX, and ImageJ) between observers (reading 2 of observer A with non *a priori* selected slices; data not shown) were found, whereas the CSMA did not significantly differ. One explanation for the differences in VAT and SAT could be the random slice selection. After all, the distribution of the intra-abdominal content (e.g., bowel) can greatly differ between slices. Consequently, single slice measurement of visceral adipose tissue would not be clinical applicable and should be reserved for clinical research of patient cohorts rather than individual patients. For SAT, the variance of subcutaneous adipose tissue distribution could have led to the observed differences. Nevertheless, all differences are relatively small and could therefore be considered as not clinically relevant The inter-observer agreement levels for OsiriX and SliceOmatic are in line with previous studies that showed a strong and significant correlation between CSMA measurements of

two observers [7, 55]. The inter-observer agreements for FatSeg and ImageJ have never been reported before, whereas a high agreement for the classification of patients with sarcopenia, as expressed in Cohen's κ, has previously been reported [7].

Several limitations apply to the current study and the used software programs. First, both observers in the current study were experienced in quantifying skeletal muscle mass using these software programs prior to conducting this study. Therefore, the agreement rates that were obtained may not apply to less experienced users. Second, OsiriX is only compatible with Macintosh, which is less commonly used in clinical practice. Furthermore, FatSeg is not freely downloadable as it is an in-house developed software program that has not been made publically available, in contrast to OsiriX and ImageJ. A license is required for the use of SliceOmatic. Third, this study could only assess the agreement of the measurement with different software programs on the same data. Intra- and inter-scanner reproducibility of the measurement could not be assessed with the current study design. Lastly, previous studies reported an approximate time of eight minutes to quantify skeletal muscle, visceral and subcutaneous adipose tissue in liver transplant patients using SliceOmatic [55]. Although some differences in user-friendliness were observed while performing the measurements, these were not objectively observed and scored in the current study. Consequently, these are not described.

In conclusion, this study showed that four different software programs have an excellent agreement to measure VAT and SAT, and CSMA in particular on abdominal CT scans, which enables reliable comparison of results of studies that use these different software programs. Multiple slice analysis is preferred for VAT and SAT measurements.

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REFERENCES

1. Leo S, Accettura C, Gnoni A, Licchetta A, Giampaglia M, Mauro A, et al. Systemic treatment of gastrointestinal cancer in elderly patients. J Gastrointest Cancer 2013; 44: 22-32.
2. Englesbe MJ, Lee JS, He K, Fan L, Schaubel DE, Sheetz KH, et al. Analytic morphomics, core muscle size, and surgical outcomes. Annals of surgery 2012; 256: 255-61.
3. Fried LP, Hadley EC, Walston JD, Newman AB, Guralnik JM, Studenski S, et al. From bedside to bench: research agenda for frailty. Science of aging knowledge environment : SAGE KE 2005; 2005: pe24.
4. Robinson TN, Wu DS, Stiegmann GV, Moss M. Frailty predicts increased hospital and six-month healthcare cost following colorectal surgery in older adults. American journal of surgery 2011; 202: 511-4.
5. Tan KY, Kawamura YJ, Tokomitsu A, Tang T. Assessment for frailty is useful for predicting morbidity in elderly patients undergoing colorectal cancer resection whose comorbidities are already optimized. American journal of surgery 2012; 204: 139-43.
6. Tegels JJ, de Maat MF, Hulsewe KW, Hoofwijk AG, Stoot JH. Value of Geriatric Frailty and Nutritional Status Assessment in Predicting Postoperative Mortality in Gastric Cancer Surgery. Journal of gastrointestinal surgery : official journal of the Society for Surgery of the Alimentary Tract 2014.
7. Reisinger KW, van Vugt JL, Tegels JJ, Snijders C, Hulsewe KW, Hoofwijk AG, et al. Functional compromise reflected by sarcopenia, frailty, and nutritional depletion predicts adverse postoperative outcome after colorectal cancer surgery. Annals of surgery 2015; 261: 345-52.
8. Rolland Y, Abellan van Kan G, Gillette-Guyonnet S, Vellas B. Cachexia versus sarcopenia. Current opinion in clinical nutrition and metabolic care 2011; 14: 15-21.
9. Cooper C, Dere W, Evans W, Kanis JA, Rizzoli R, Sayer AA, et al. Frailty and sarcopenia: definitions and outcome parameters. Osteoporos Int 2012; 23: 1839-48.
10. Marzetti E, Leeuwenburgh C. Skeletal muscle apoptosis, sarcopenia and frailty at old age. Exp Gerontol 2006; 41: 1234-8.
11. Roubenoff R. Sarcopenia: a major modifiable cause of frailty in the elderly. J Nutr Health Aging 2000; 4: 140-2.
12. Baumgartner RN, Koehler KM, Gallagher D, Romero L, Heymsfield SB, Ross RR, et al. Epidemiology of sarcopenia among the elderly in New Mexico. Am J Epidemiol 1998; 147: 755-63.
13. Fearon K, Strasser F, Anker SD, Bosaeus I, Bruera E, Fainsinger RL, et al. Definition and classification of cancer cachexia: an international consensus. Lancet Oncol 2011; 12: 489-95.
14. Evans WJ. Skeletal muscle loss: cachexia, sarcopenia, and inactivity. The American journal of clinical nutrition 2010; 91: 1123S-7S.

15. Evans WJ, Morley JE, Argiles J, Bales C, Baracos V, Guttridge D, et al. Cachexia: a new definition. Clin Nutr 2008; 27: 793-9.

16. Bachmann J, Heiligensetzer M, Krakowski-Roosen H, Buchler MW, Friess H, Martignoni ME. Cachexia worsens prognosis in patients with resectable pancreatic cancer. Journal of gastrointestinal surgery : official journal of the Society for Surgery of the Alimentary Tract 2008; 12: 1193-201.

17. Antoun S, Baracos VE, Birdsell L, Escudier B, Sawyer MB. Low body mass index and sarcopenia associated with dose-limiting toxicity of sorafenib in patients with renal cell carcinoma. Annals of oncology : official journal of the European Society for Medical Oncology / ESMO 2010; 21: 1594-8.

18. Prado CM, Birdsell LA, Baracos VE. The emerging role of computerized tomography in assessing cancer cachexia. Curr Opin Support Palliat Care 2009; 3: 269-75.

19. Fearon KC. Cancer cachexia: developing multimodal therapy for a multidimensional problem. Eur J Cancer 2008; 44: 1124-32.

20. Tisdale MJ. Mechanisms of cancer cachexia. Physiological reviews 2009; 89: 381-410.

21. Acharyya S, Butchbach ME, Sahenk Z, Wang H, Saji M, Carathers M, et al. Dystrophin glycoprotein complex dysfunction: a regulatory link between muscular dystrophy and cancer cachexia. Cancer cell 2005; 8: 421-32.

22. Loberg RD, Bradley DA, Tomlins SA, Chinnaiyan AM, Pienta KJ. The lethal phenotype of cancer: the molecular basis of death due to malignancy. CA: a cancer journal for clinicians 2007; 57: 225-41.

23. Prado CM, Lieffers JR, McCargar LJ, Reiman T, Sawyer MB, Martin L, et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. Lancet Oncol 2008; 9: 629-35.

24. Huillard O, Mir O, Peyromaure M, Tlemsani C, Giroux J, Boudou-Rouquette P, et al. Sarcopenia and body mass index predict sunitinib-induced early dose-limiting toxicities in renal cancer patients. Br J Cancer 2013; 108: 1034-41.

25. Antoun S, Borget I, Lanoy E. Impact of sarcopenia on the prognosis and treatment toxicities in patients diagnosed with cancer. Curr Opin Support Palliat Care 2013; 7: 383-9.

26. Levolger S, van Vugt JL, de Bruin RW, IJzermans JN. Systematic review of sarcopenia in patients operated on for gastrointestinal and hepatopancreatobiliary malignancies. Br J Surg 2015; 102: 1448-58.

27. Lee JS, He K, Harbaugh CM, Schaubel DE, Sonnenday CJ, Wang SC, et al. Frailty, core muscle size, and mortality in patients undergoing open abdominal aortic aneurysm repair. J Vasc Surg 2011; 53: 912-7.

28. Carey EJ. Sarcopenia in solid organ transplantation. Nutr Clin Pract 2014; 29: 159-70.

29. van Vugt JL, Levolger S, de Bruin RW, van Rosmalen J, Metselaar HJ, IJzermans JN. Systematic review and meta-analysis of the impact of computed tomography assessed skeletal muscle mass on outcome in patients awaiting or undergoing liver transplantation. Am J Transplant 2016.

30. Cakir H, Heus C, van der Ploeg TJ, Houdijk AP. Visceral obesity determined by CT scan and outcomes after colorectal surgery; a systematic review and meta-analysis. Int J Colorectal Dis 2015; 30: 875-82.

31. Cecchini S, Cavazzini E, Marchesi F, Sarli L, Roncoroni L. Computed tomography volumetric fat parameters versus body mass index for predicting short-term outcomes of colon surgery. World J Surg 2011; 35: 415-23.

32. Jung M, Volonte F, Buchs NC, Gayet-Ageron A, Pugin F, Gervaz P, et al. Perirenal fat surface area as a risk factor for morbidity after elective colorectal surgery. Dis Colon Rectum 2014; 57: 201-9.

33. van Vledder MG, Levolger S, Ayez N, Verhoef C, Tran TC, IJzermans JN. Body composition and outcome in patients undergoing resection of colorectal liver metastases. Br J Surg 2012; 99: 550-7.

34. Doyle SL, Bennett AM, Donohoe CL, Mongan AM, Howard JM, Lithander FE, et al. Establishing computed tomography-defined visceral fat area thresholds for use in obesity-related cancer research. Nutr Res 2013; 33: 171-9.

35. Mitsiopoulos N, Baumgartner RN, Heymsfield SB, Lyons W, Gallagher D, Ross R. Cadaver validation of skeletal muscle measurement by magnetic resonance imaging and computerized tomography. J Appl Physiol (1985) 1998; 85: 115-22.

36. Yoshizumi T, Nakamura T, Yamane M, Islam AH, Menju M, Yamasaki K, et al. Abdominal fat: standardized technique for measurement at CT. Radiology 1999; 211: 283-6.

37. Gomez-Perez SL, Haus JM, Sheean P, Patel B, Mar W, Chaudhry V, et al. Measuring Abdominal Circumference and Skeletal Muscle From a Single Cross-Sectional Computed Tomography Image: A Step-by-Step Guide for Clinicians Using National Institutes of Health ImageJ. JPEN J Parenter Enteral Nutr 2016; 40: 308-18.

38. Bland JM, Altman DG. Statistical methods for assessing agreement between two methods of clinical measurement. Lancet 1986; 1: 307-10.

39. Shrout PE, Fleiss JL. Intraclass correlations: uses in assessing rater reliability. Psychol Bull 1979; 86: 420-8.

40. Jaccard P. Étude comparative de la distribution florale dans une portion des alpes et des jura. Bulletin de la Société Vaudoise des Sciences Naturelles 1901; 37: 547-79.

41. Sharma P, Zargar-Shoshtari K, Caracciolo JT, Richard GJ, Poch MA, Pow-Sang J, et al. Sarcopenia as a predictor of complications in penile cancer patients undergoing inguinal lymph node dissection. World J Urol 2015.

42. Wan F, Zhu Y, Gu C, Yao X, Shen Y, Dai B, et al. Lower skeletal muscle index and early complications in patients undergoing radical cystectomy for bladder cancer. World J Surg Oncol 2014; 12: 14.

43. Miller BS, Ignatoski KM, Daignault S, Lindland C, Doherty M, Gauger PG, et al. Worsening central sarcopenia and increasing intra-abdominal fat correlate with decreased survival in patients with adrenocortical carcinoma. World J Surg 2012; 36: 1509-16.

44. Kuroki LM, Mangano M, Allsworth JE, Menias CO, Massad LS, Powell MA, et al. Pre-operative Assessment of Muscle Mass to Predict Surgical Complications and Prognosis in Patients With Endometrial Cancer. Ann Surg Oncol 2014.

45. Fairchild B, Webb TP, Xiang Q, Tarima S, Brasel KJ. Sarcopenia and Frailty in Elderly Trauma Patients. World J Surg 2014.

46. Mir O, Coriat R, Blanchet B, Durand JP, Boudou-Rouquette P, Michels J, et al. Sarcopenia predicts early dose-limiting toxicities and pharmacokinetics of sorafenib in patients with hepatocellular carcinoma. PLoS One 2012; 7: e37563.

47. Collins J, Noble S, Chester J, Coles B, Byrne A. The assessment and impact of sarcopenia in lung cancer: a systematic literature review. BMJ Open 2014; 4: e003697.

48. Sabel MS, Lee J, Cai S, Englesbe MJ, Holcombe S, Wang S. Sarcopenia as a prognostic factor among patients with stage III melanoma. Ann Surg Oncol 2011; 18: 3579-85.

49. Englesbe MJ, Patel SP, He K, Lynch RJ, Schaubel DE, Harbaugh C, et al. Sarcopenia and mortality after liver transplantation. Journal of the American College of Surgeons 2010; 211: 271-8.

50. Irving BA, Weltman JY, Brock DW, Davis CK, Gaesser GA, Weltman A. NIH ImageJ and Slice-O-Matic computed tomography imaging software to quantify soft tissue. Obesity (Silver Spring) 2007; 15: 370-6.

51. Wu GA, Bogie K. Assessment of gluteus maximus muscle area with different image analysis programs. Arch Phys Med Rehabil 2009; 90: 1048-54.

52. Strandberg S, Wretling ML, Wredmark T, Shalabi A. Reliability of computed tomography measurements in assessment of thigh muscle cross-sectional area and attenuation. BMC Med Imaging 2010; 10: 18.

53. Dello SA, Stoot JH, van Stiphout RS, Bloemen JG, Wigmore SJ, Dejong CH, et al. Prospective volumetric assessment of the liver on a personal computer by nonradiologists prior to partial hepatectomy. World J Surg 2011; 35: 386-92.

54. Richards CH, Roxburgh CS, MacMillan MT, Isswiasi S, Robertson EG, Guthrie GK, et al. The relationships between body composition and the systemic inflammatory response in patients with primary operable colorectal cancer. PLoS One 2012; 7: e41883.

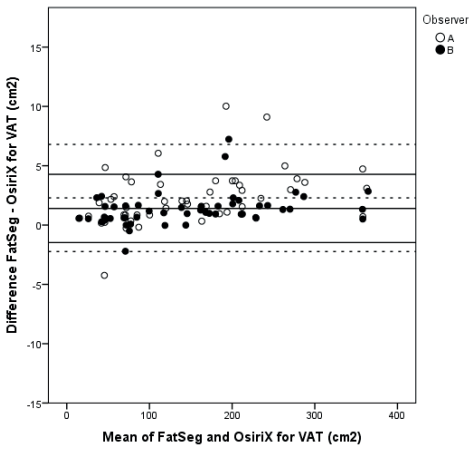
55. Cruz RJ, Jr., Dew MA, Myaskovsky L, Goodpaster B, Fox K, Fontes P, et al. Objective radiologic assessment of body composition in patients with end-stage liver disease: going beyond the BMI. Transplantation 2013; 95: 617-22.

56. Fortin M, Battie MC. Quantitative paraspinal muscle measurements: inter-software reliability and agreement using OsiriX and ImageJ. Phys Ther 2012; 92: 853-64.

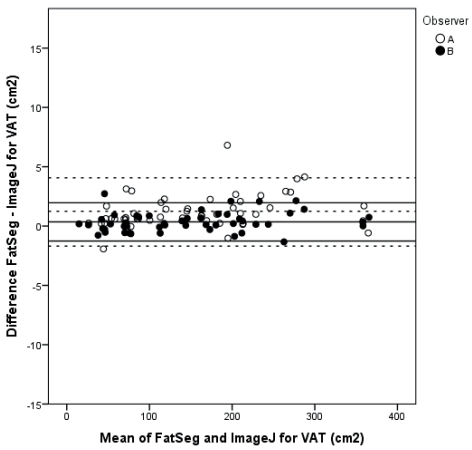
57. Shen W, Punyanitya M, Wang Z, Gallagher D, St-Onge MP, Albu J, et al. Total body skeletal muscle and adipose tissue volumes: estimation from a single abdominal cross-sectional image. J Appl Physiol (1985) 2004; 97: 2333-8.

SUPPLEMENTARY MATERIAL

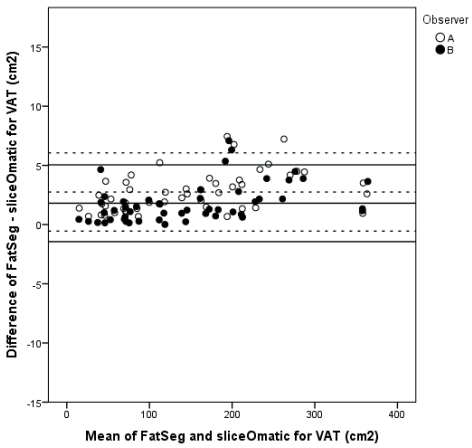
Supplementary figure 1. Bland-Altman 95% limits of agreement plots for the agreement between the various software programs (provided on the X-axes and Y-axes) for VAT (cm^2). The dotted lines are the mean of the difference and the 95% limits of agreement (± 2 SD) between the VAT of reading 1 of observer A and the solid lines of reading 1 of observer B.



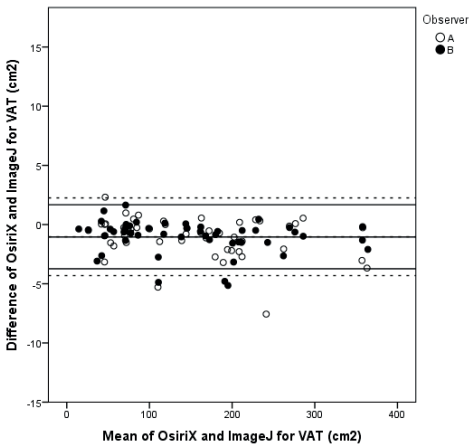
Supplementary figure 1a. There was proportional systematic bias for both observers (p=0.004 and p=0.043, respectively).



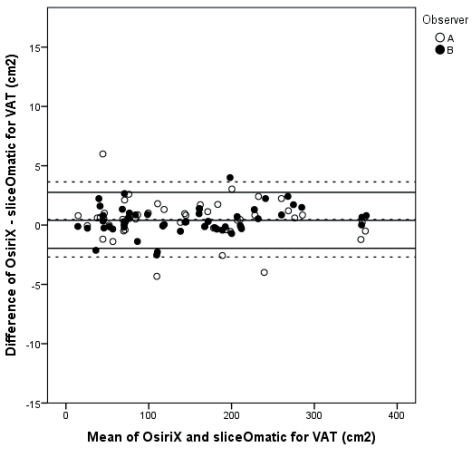
Supplementary figure 1b. There was proportional systematic bias for observer A (p=0.038), but not for observer B (p=0.154).



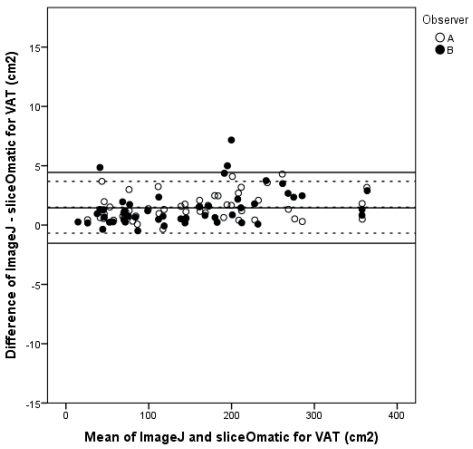
Supplementary figure 1c. There was proportional systematic bias for both observers (both p=0.002).



Supplementary figure 1d. There was proportional systematic bias for observer A (p=0.045), whereas there was no proportional systematic bias for observer B (p=0.202).

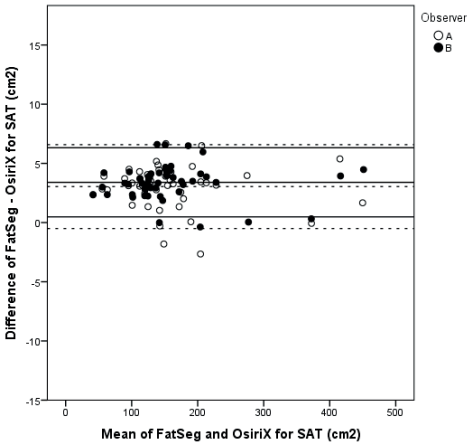


Supplementary figure 1e. There was no proportional systematic bias for any observer (p=0.412 and p=0.114, respectively).

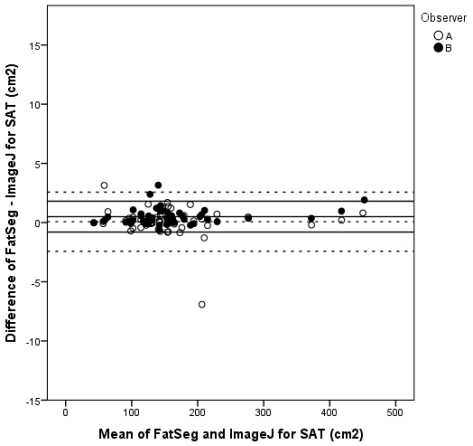


Supplementary figure 1f. There was no proportional systematic bias for observer A (p=0.068), whereas there was significant bias for observer B (p=0.014).

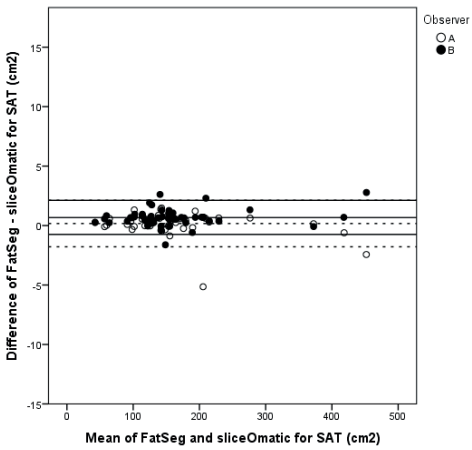
Supplementary figure 2. Bland-Altman 95% limits of agreement plots for the agreement between the various software programs (provided on the X-axes and Y-axes) for SAT (cm^2). The dotted lines are the mean of the difference and the 95% limits of agreement (± 2 SD) between the SAT of reading 1 of observer A and the solid lines of reading 1 of observer B.



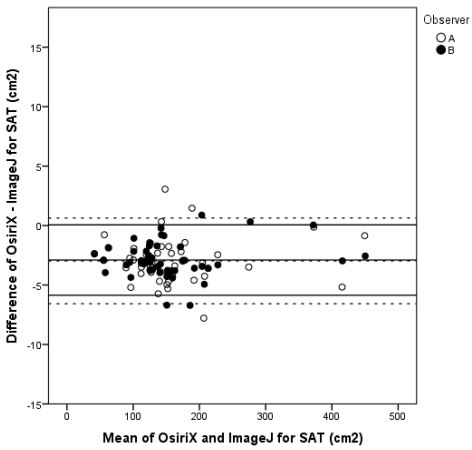
Supplementary figure 2a. There was no proportional systematic bias for any observer (p=0.534 and p=0.801, respectively).



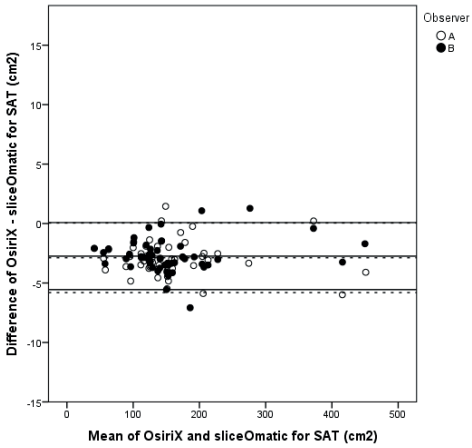
Supplementary figure 2b. There was no proportional systematic bias for any observer (p=0.538 and p=0.112, respectively).



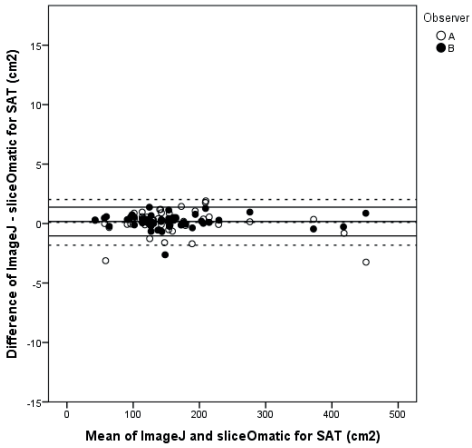
Supplementary figure 2c. There was proportional systematic bias for observer A (p=0.031), whereas there was no proportional systematic bias for observer B (p=0.134).



Supplementary figure 2d. There was no proportional systematic bias for any observer (p=0.853 and p=0.344, respectively).

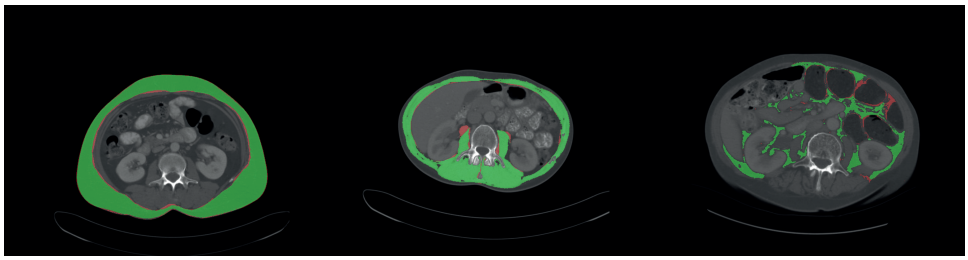


Supplementary figure 2e. There was no proportional systematic bias for any observer (p=0.511 and p=0.305, respectively).



Supplementary figure 2f. There was no proportional systematic bias for any observer (p=0.175 and p=0.939, respectively).

Supplementary figure 3. Jaccard similarity coefficients (lowest and highest are shown) for inter-observer comparisons of CSMA, VAT, and SAT (cm^2) measurements (reading 1 of observer A versus reading 1 of observer B). The green area represents similarity, whereas the red area represents discrepancy in measurements.

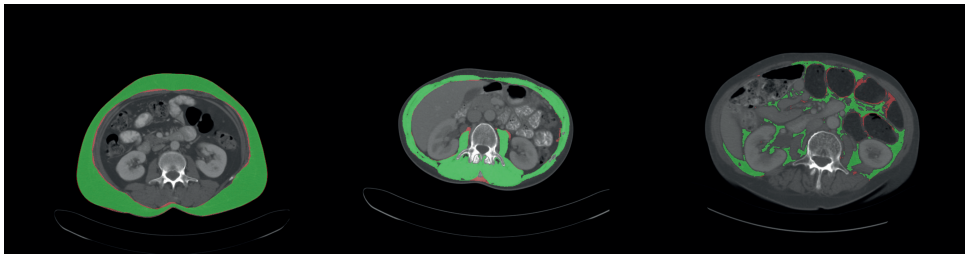


Supplementary figure 3a. The CSMA measured with FatSeg (1) and ImageJ (2), resulting in Jaccard similarity coefficients of 0.997 and 0.931, respectively.

Supplementary figure 3b. The VAT measured with FatSeg (1) and OsiriX (2), resulting in Jaccard similarity coefficients of 0.998 and 0.835, respectively.

Supplementary figure 3c. The SAT measured with OsiriX (1) and ImageJ (2), resulting in Jaccard similarity coefficients of 1.000 and 0.899, respectively.

Supplementary figure 4. Jaccard similarity coefficients (lowest and highest are shown) for intra-observer comparisons of CSMA, VAT, and SAT (cm^2) measurements (reading 1 versus reading 2 of observer A). The green area represents similarity, whereas the red area represents discrepancy in measurements.

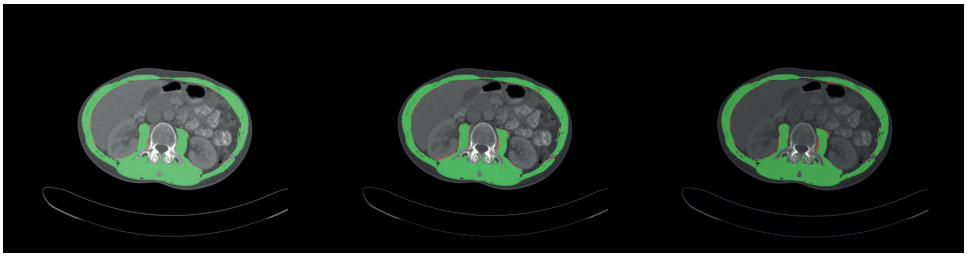


Supplementary figure 4a. The CSMA measured with FatSeg (1) and OsiriX (2), resulting in Jaccard similarity coefficients of 1.000 and 0.953, respectively.

Supplementary figure 4b. The VAT measured with FatSeg (1) and OsiriX (2), resulting in Jaccard similarity coefficients of 0.999 and 0.838, respectively.

Supplementary figure 4c. The SAT measured with ImageJ (1 and 2), resulting in Jaccard similarity coefficients of 1.000 and 0.900, respectively. Furthermore, a Jaccard similarity coefficient of 1.000 was also observed in two other patients measured with FatSeg and OsiriX.

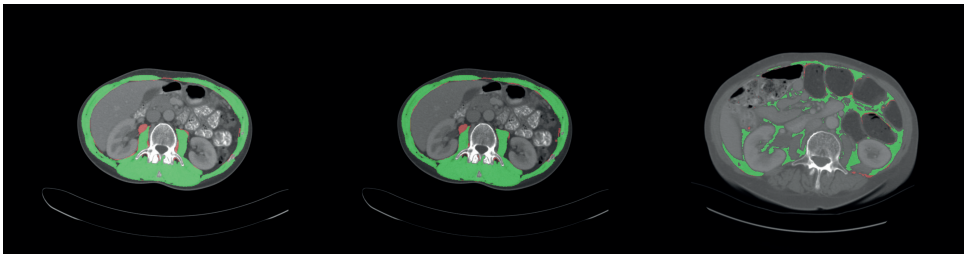
Supplementary figure 5. All worst Jaccard similarity coefficients for the inter-software (5a), inter-observer (5b) and intra-observer (5c) agreement for the three body composition analyses except the ones that are shown in figure 2 and supplementary figures 2 and 3.



Supplementary figure 5a1a. CSMA measured with FatSeg and Image resulting in a Jaccard similarity coefficient of 0.959.

Supplementary figure 5a1b. CSMA measured with FatSeg and OsiriX resulting in a Jaccard similarity coefficient of 0.948.

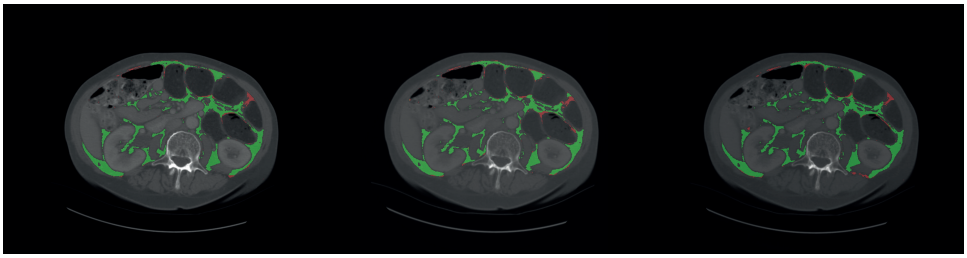
Supplementary figure 5a1c. CSMA measured with ImageJ and OsiriX resulting in a Jaccard similarity coefficient of0.948.



Supplementary figure 5a1d. CSMA measured with ImageJ and SliceOmatic resulting in a Jaccard similarity coefficient of 0.932.

Supplementary figure 5a1e. CSMA measured with OsiriX and SliceOmatic resulting in a Jaccard similarity coefficient of0.944.

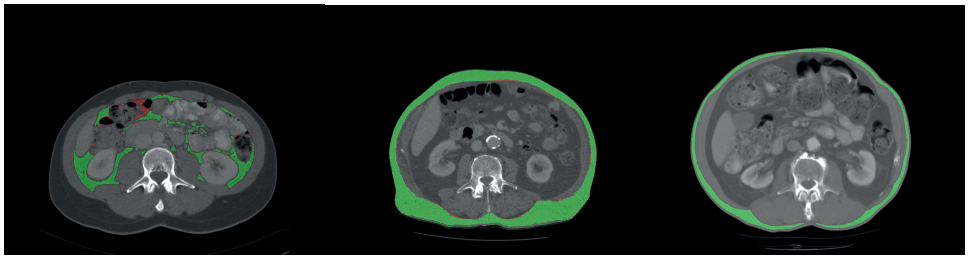
Supplementary figure 5a2a. VAT measured with FatSeg and ImageJ resulting in a Jaccard similarity coefficient of0.903.



Supplementary figure 5a2b. VAT measured with FatSeg and OsiriX resulting in a Jaccard similarity coefficient of 0.886.

Supplementary figure 5a2c. VAT measured with FatSeg and SliceOmatic resulting in a Jaccard similarity coefficient of 0.860.

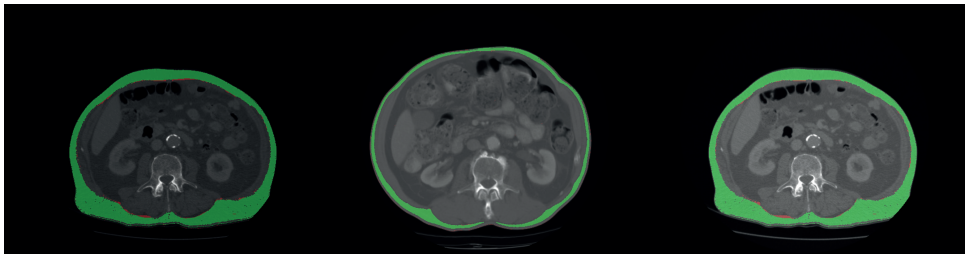
Supplementary figure 5a2d. VAT measured with ImageJ and OsiriX resulting in a Jaccard similarity coefficient of 0.891.



Supplementary figure 5a2e. VAT measured with OsiriX and SliceOmatic resulting in a Jaccard similarity coefficient of 0.884.

Supplementary figure 5a3a. SAT measured with FatSeg and ImageJ resulting in a Jaccard similarity coefficient of0.968.

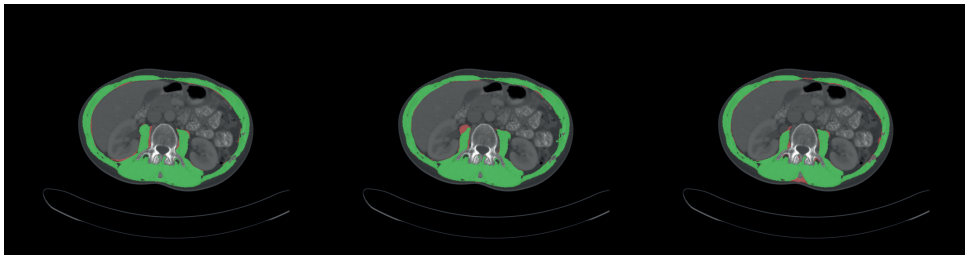
Supplementary figure 5a3b. SAT measured with FatSeg and OsiriX resulting in a Jaccard similarity coefficient of 0.926.



Supplementary figure 5a3c. SAT measured with FatSeg and SliceOmatic resulting in a Jaccard similarity coefficient of 0.960.

Supplementary figure 5a3d. SAT measured with ImageJ and OsiriX resulting in a Jaccard similarity coefficient of 0.927.

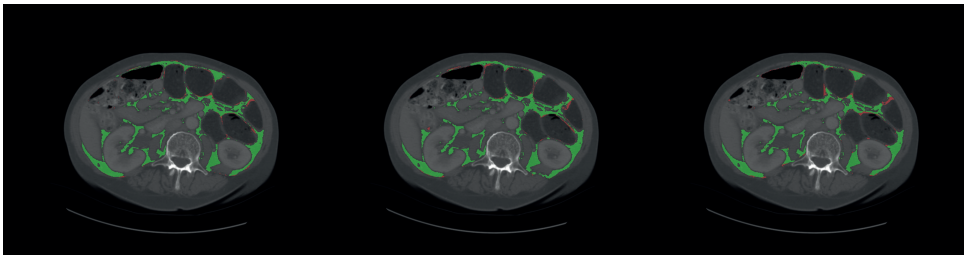
Supplementary figure 5a3e. SAT measured with ImageJ and SliceOmatic resulting in a Jaccard similarity coefficient of 0.965.



Supplementary figure 5b1a. CSMA measured with FatSeg resulting in a Jaccard similarity coefficient of 0.949.

Supplementary figure 5b1b. CSMA measured with OsiriX resulting in a Jaccard similarity coefficient of 0.931.

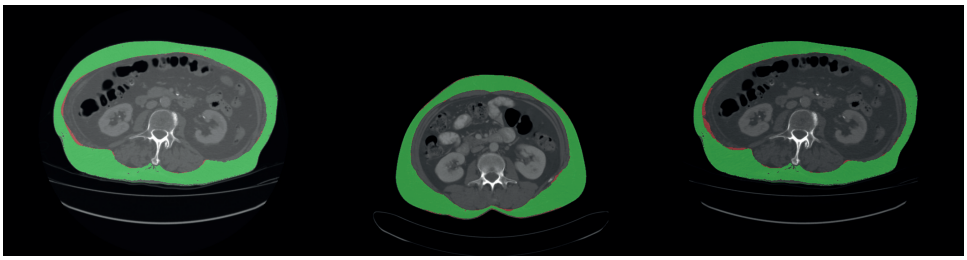
Supplementary figure 5b1c. CSMA measured with SliceOmatic resulting in a Jaccard similarity coefficient of 0.939.



Supplementary figure 5b2a. VAT measured with FatSeg resulting in a Jaccard similarity coefficient of 0.908.

Supplementary figure 5b2b. VAT measured with ImageJ resulting in a Jaccard similarity coefficient of 0.905.

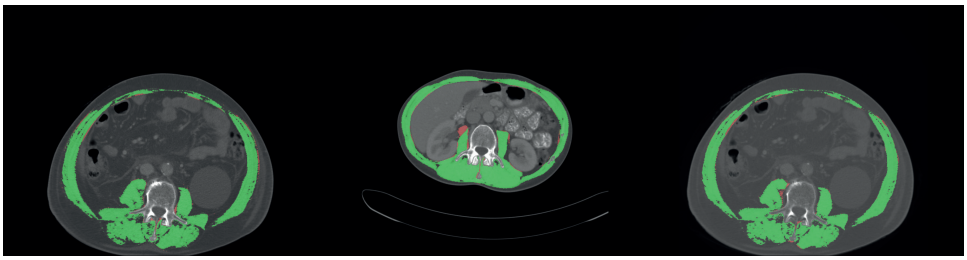
Supplementary figure 5b2c. VAT measured with SliceOmatic resulting in a Jaccard similarity coefficient of 0.876.



Supplementary figure 5b3a. SAT measured with FatSeg resulting in a Jaccard similarity coefficient of 0.969.

Supplementary figure 5b3b. SAT measured with OsiriX resulting in a Jaccard similarity coefficient of 0.961.

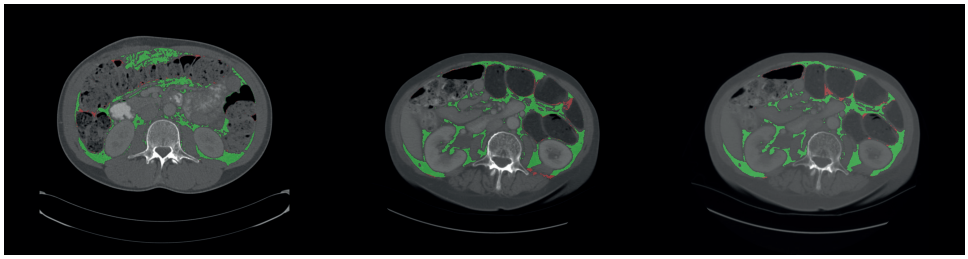
Supplementary figure 5b3c. SAT measured with SliceOmatic resulting in a Jaccard similarity coefficient of 0.959.



Supplementary figure 5c1a. CSMA measured with FatSeg resulting in a Jaccard similarity coefficient of 0.961.

Supplementary figure 5c1b. CSMA measured with ImageJ resulting in a Jaccard similarity coefficient of 0.948.

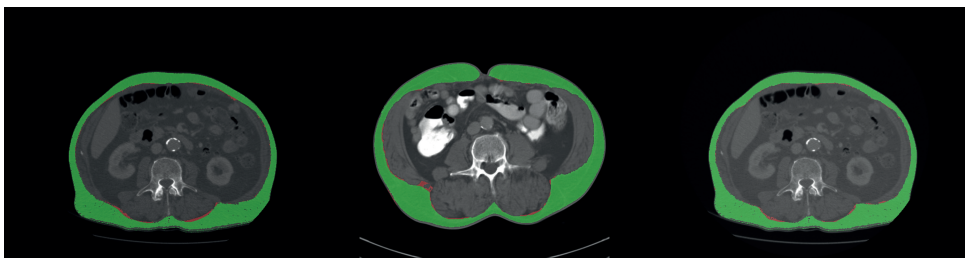
Supplementary figure 5c1c. CSMA measured with SliceOmatic resulting in a Jaccard similarity coefficient of 0.961.



Supplementary figure 5c2a. VAT measured with FatSeg resulting in a Jaccard similarity coefficient of 0.916.

Supplementary figure 5c2b. VAT measured with ImageJ resulting in a Jaccard similarity coefficient of 0.891.

Supplementary figure 5c2c. VAT measured with SliceOmatic resulting in a Jaccard similarity coefficient of 0.901.



Supplementary figure 5c3a. SAT measured with FatSeg resulting in a Jaccard similarity coefficient of 0.956.

Supplementary figure 5c3b. SAT measured with OsiriX resulting in a Jaccard similarity coefficient of 0.967.

Supplementary figure 5c3c. SAT measured with SliceOmatic resulting in a Jaccard similarity coefficient of 0.967.

CHAPTER 3

Contrast-Enhancement Influences Skeletal Muscle Density, but not Skeletal Muscle Mass, Measurements on Computed Tomography

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ABSTRACT

Background: Low skeletal muscle mass and density have recently been discovered as prognostic and predictive parameters to guide interventions in various populations, including cancer patients. The gold standard for body composition analysis in cancer patients is computed tomography (CT). To date, the effect of contrast-enhancement on muscle composition measurements has not been established. The aim of this study was to determine the effect of contrast-enhancement on skeletal muscle mass and density measurements on four-phase CT studies.

Methods: In this observational study, two observers measured cross-sectional skeletal muscle area corrected for patients' height (skeletal muscle index [SMI]) and density (SMD) at the level of the third lumbar vertebra on 50 randomly selected CT-examinations with unenhanced, arterial, and portal-venous phases. The levels of agreement between enhancement phases for SMI and SMD were calculated using intra-class correlation coefficients (ICCs).

Results: Mean SMI was 42.5 (±9.9) cm^2/m^2 on the unenhanced phase, compared with 42.8 (±9.9) and 43.6 (±9.9) cm^2/m^2 for the arterial and portal-venous phase, respectively (both $p<0.01$). Mean SMD was lower for the unenhanced phase (30.9±8.0 Hounsfield Units [HU]) compared with the arterial (38.0±9.9 HU) and portal-venous (38.7±9.2 HU) phase (both $p<0.001$). No significant difference was found between SMD in the portal-venous and arterial phase ($p=0.161$). The ICCs were excellent (≥0.992) for all SMIs and for SMD between the contrast-enhanced phases (0.949). The ICCs for the unenhanced phase compared with the arterial (0.676) and portal-venous (0.665) phase were considered fair to good.

Conclusions: Statistically significant differences in SMI were assessed between different enhancement phases. However, further work is needed to assess the clinical relevance of these small differences. Contrast-enhancement strongly influenced SMD values. Studies using this measure should therefore use the portal-venous phase of contrast-enhanced CT-examinations.

INTRODUCTION

The involuntary loss of skeletal muscle mass, quality and function is considered to be a result of aging (i.e. sarcopenia), or as part of muscle wasting syndromes (i.e. cancer cachexia, chronic diseases, bed rest) [1-3]. Low skeletal muscle mass has recently been identified as a prognostic factor for treatment outcome and survival in various populations, such as in cancer and liver transplant patients [4, 5]. Furthermore, it is associated with an increased risk of postoperative complications, chemotherapy toxicity and increased hospital expenditure [4, 6-8]. Low skeletal muscle density, a measure for intramuscular adipose content and muscle quality, has recently been described as a risk factor for mortality in patients with lymphoma, melanoma, metastatic renal cell carcinoma, pancreatic carcinoma, and metastatic gastric cancer [9-13]. Body composition measures may guide future interventions to manage skeletal muscle wasting and to increase patients' resistance towards stressors, such as surgery and chemotherapy [14].

The gold standard and most used modality to assess body composition is computed tomography (CT) due to its wide availability, especially in cancer patients [15-17]. Excellent inter-observer and intra-observer agreement, as well as excellent comparability of various commonly used software programs for skeletal muscle mass measurement have previously been described [18]. However, the effect of contrast-enhancement on skeletal muscle mass and density measurements remains unclear. It is well-known that contrast-enhancement may influence tissue attenuation [19] and may consequently influence skeletal muscle mass and density measurements. Nevertheless, various enhancement phases have been used in studies that investigated the association between CT-assessed skeletal muscle mass and density and outcome measures [9-12, 20]. Therefore, the aim of this study was to compare skeletal muscle mass and density measurements on CT between different contrast-enhancement phases.

METHODS

Patients

A total of 50 patients with cancer or evaluated for liver transplantation in Erasmus MC University Medical Center between 2009 and 2015 with available multiphase (unenhanced, arterial, portal-venous) abdominal CT-examinations were randomly selected retrospectively. Patients with CTs on which part of the cross-sectional skeletal muscle area was not depicted (e.g., due to obesity) or with artefacts (e.g., due to prostheses) were excluded. Date of birth, sex, body weight, and body height were collected from the electronic patient files within a month of the CT-examination. Body mass index (BMI) was calculated and patients were categorized as underweight (BMI <18.5), normal weight (BMI 20.0-24.9), overweight (BMI 25.0-29.9) or obese (BMI ≥30.0) according to the World Health Organization (WHO) definitions [21]. Approval from the local medical ethical committee was obtained and the study has been performed according to the 1964 Declaration of Helsinki and its later amendments.

CT scanning protocol

All CT examinations were performed according to a standardised protocol. First, an unenhanced phase was obtained. Afterwards, intravenous (IV) contrast administration in an antecubital vein followed by saline flush of 20ml was performed using a power injector. The contrast material used was Visipaque 320 mgI/ml (GE Healthcare, Cork, Ireland), adapted to a patient's body weight. Patients with body weight <80 kg received 120 ml contrast medium, whereas patients with body weight ≥80 kg received 150 ml contrast medium. Phases acquired were the arterial phase, determined using a bolus-tracking technique, followed by the portal-venous phase acquired 70 seconds after contrast administration. For the arterial phase, a region of interest (ROI) is placed in the upper abdominal aorta; when the threshold of +100 HU is reached, scanning starts with a delay of 15 seconds. Estimated time after administration of the bolus is 30-35 seconds for the arterial phase. The portal-venous phase is obtained with a fixed delay of 70 seconds after administration of the contrast material. Axial reconstructions were created with a slice thickness of 3mm in all phases. No adverse reactions were noted during contrast administration. All images were transferred to our local picture archiving and communication system (PACS).

An experienced abdominal radiologist (FEJAW) confirmed the different phases of contrast-enhancement per patient. Furthermore, the mean intraluminal attenuation (in HU) of the aorta was measured for every phase per patient.

Skeletal muscle mass and density measurements

The cross sectional muscle area (CSMA) was measured at the level of the third lumbar vertebra for the various contrast-enhancement phases (i.e. unenhanced, arterial, portal-venous). The selected slice was the one on which both transversal processes were visible. Two observers (HJWS and KMV) who were blinded for patient characteristics performed all measurements as previously described [22]. An in-house developed software program (FatSeg, developed by the Biomedical Imaging Group Rotterdam of Erasmus MC, Rotterdam, The Netherlands, using MeVisLab [Mevis Medical Solutions, Bremen, Germany]) was used (figure 1). A previous study indicated excellent comparability between this and other frequently used software programs (i.e. SliceOmatic, OsiriX, and ImageJ) for body composition analyses [23]. The inner and outer contours of the CSMA (including the psoas, rectus abdominis, transversus abdominis, internal and external abdominal oblique muscles) were manually outlined and the tissue within the threshold of -30 to +150 Hounsfield units (HU) was selected. CSMA was corrected for patients' body height squared, as is common for body composition measures, resulting in the skeletal muscle index (SMI, cm^2/m^2). The mean HU value was recorded as a measure of skeletal muscle density. Low skeletal muscle mass was defined using previously described cut-off values: skeletal muscle index <41 cm^2/m^2 for women regardless of BMI, and <43 and <53 cm^2/m^2 for men with BMI <25.0 and ≥ 25.0 kg/m^2, respectively. The definition for low skeletal muscle density was identical for men and women: a skeletal muscle attenuation <41 for patients with BMI <25.0 kg/m^2 and <33 for patients with BMI ≥ 25.0 kg/m^2 [24].

Statistical analysis

Normality of data was tested using the Shapiro-Wilk test. Continuous data are presented as median with interquartile range (IQR) or mean with standard deviation (±), depending on the normality of distribution. Categorical data are presented as counts with percentages. Differences between the different contrast-enhancement phases were tested using a paired t-test or Wilcoxon signed rank test, again depending on the normality of the distribution of the data. The agreement between observers (i.e. inter-observer agreement) and between contrast-enhancement phases (i.e. inter-enhancement phase agreement) were calculated using intra-class correlation coefficients (ICCs) with 95% confidence intervals (CIs) using a two-way mixed single measures model with absolute agreement. Bland and Altman plots with 95% CI were generated to investigate the agreement between contrast-enhancement phases [25]. Linear regression was used to test for proportional systematic bias [26]. Smallest detectable changes (SDC), expressing the smallest detectable difference considered a ''real'' change in paired measures, were calculated for both skeletal muscle mass and density for the different contrast-enhancement phases using the following formula:

$$Change\ [in\ skeletal\ muscle\ mass\ or\ density] \pm (1.96\ x\ \frac{SEMchange}{\sqrt{k}}),$$

in which k is the number of measurements and SEM stands for standard error of the measurement [27]. The SEM is calculated using the following formula:

$$Standard\ Deviation\ [SD]\ x\ \sqrt{1 - ICC}.$$

The agreement on sarcopenia assessment between observers and contrast-enhancement phases was calculated using Cohen's κ coefficients. ICCs and Cohen's κ's ranging from 0.00 to 0.49 were interpreted was poor, whereas coefficients ranging from 0.50 to 0.74 and 0.75 to 1.00 were interpreted as fair to good and excellent, respectively [28].

The average of the two measurements by the two observers was used. Two-sided p-values <0.05 were considered statistically significant. All statistical analyses were performed using SPSS for Windows (IBM Corp., Armonk, NY, USA), version 22.

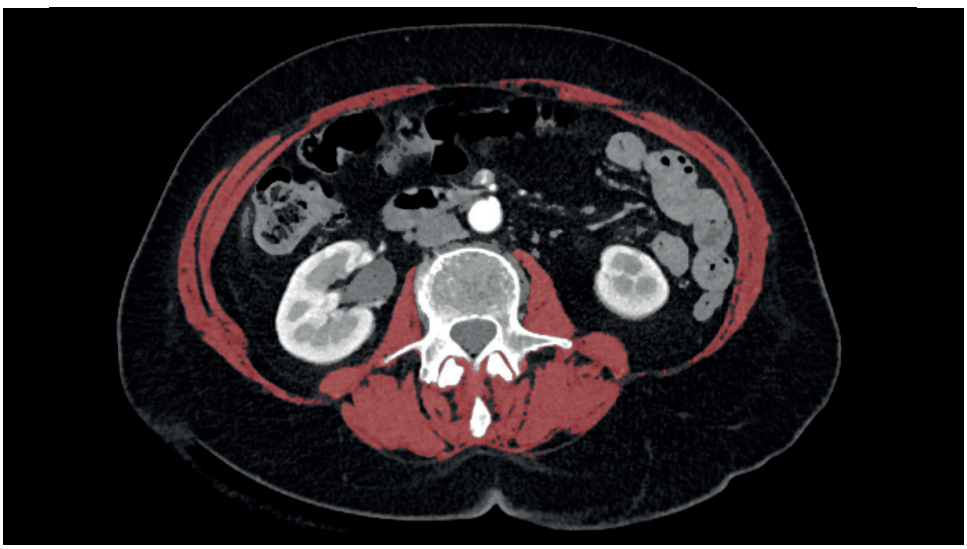


Figure 1. Example of skeletal muscle mass and density measurement on a contrast-enhanced CT slice in the portal-venous phase at the level of the third lumbar vertebra (L3). The cross-sectional skeletal muscle area of this 71-year-old woman with a body mass index of 24.7 kg/m^2 was 95.6 cm^2, resulting in a skeletal muscle index of 33.1 cm^2/m^2. The mean skeletal muscle attenuation was 33 Hounsfield units. According to the cut-off values of Martin *et al.*, this patient is considered to have both sarcopenia and low skeletal muscle density.

RESULTS

Patient and CT characteristics

The study cohort consisted of 23 (46%) females and 27 (54%) males with a mean BMI of 24.2 (± 4.0) kg/m^2. In total, 19 (38%) patients had a BMI ≥25 kg/m^2 and 4 (8%) patients were considered obese (i.e. BMI ≥30 kg/m^2). All baseline characteristics are shown in supplementary table 1. The median intraluminal attenuation of the aorta was 41 (IQR 37-45) HU in CT images without contrast-enhancement, 404 (IQR 320-514) HU in the arterial contrast-enhancement phase, and 158 (IQR 143-189) HU in the venous contrast-enhancement phase (figure 2). The inter-observer ICCs were 0.999 for all contrast-enhancement phases for the skeletal muscle area, and ≥0.980 for the skeletal muscle density.

Skeletal muscle mass measurements

An overall difference in SMI was found between the three contrast-enhancement phases ($F(2, 98) = 56.174$, $p<0.001$). The mean skeletal muscle index was 42.5 ± 9.9 cm^2/m^2 on the unenhanced phase, which was significantly lower compared with the arterial phase (42.8 ± 9.9 cm^2/m^2, p=0.021) and the portal-venous phase (43.6 ± 9.9 cm^2/m^2, p<0.001) (table 1, figure 3). A significant difference was also observed between the arterial and portal-venous phase (42.8 versus 43.6 cm^2/m^2, p<0.001). Bland Altman plots with 95% limits of agreement for the SMI are shown in figure 4. There was no proportional systematic bias for any comparison. The ICCs were excellent (all >0.99) for all comparisons (table 1). Comparable results were found when using the cross-sectional muscle area (CSMA, supplementary figure 1). According to the cut-off values defined by Martin *et al.* [24], 22 (44%) patients were considered to have low skeletal muscle mass using unenhanced CT, compared with 21 (42%) patients using the arterial phase and 24 (48%) patients using the portal-venous phase. This resulted in excellent Cohen's κ's of 0.959 (unenhanced versus arterial phase), 0.920 (unenhanced versus portal-venous phase), and 0.879 (arterial versus portal-venous phase) (table 2).

Skeletal muscle density measurements

An overall significant difference in skeletal muscle density between the three contrast-enhancement phases was found ($F(1.649, 80.813) = 150.167$, $p<0.001$). The mean skeletal muscle density was lower for the unenhanced phase (30.9 ± 8.0 HU) compared with the arterial (38.0 ± 9.9 HU) and portal-venous (38.7 ± 9.2 HU) phase (both p<0.001), but not between the two latter (38.0 versus 38.7 HU, p=0.483) (table 1, figure 5).

Table 1. Differences in skeletal muscle mass and skeletal muscle density measurements using unenhanced, arterial, and portal-venous phases on CT.

	Skeletal muscle area (cm^2)					Skeletal muscle mass (cm^2/m^2)					Skeletal muscle density (HU)				
	Mean difference (cm^2)	p-value	ICC	SEM (cm^2)	SDC (cm^2)	Mean difference (cm^2/m^2)	p-value	ICC	SEM (cm^2/m^2)	SDC (cm^2/m^2)	Mean difference (HU)	p-value	ICC	SEM (HU)	SDC (HU)
Unenhanced - Arterial phase	-0.9	*0.011*	0.997 (0.994-0.998)	0.14	0.38	-0.3	0.007	0.996 (0.992-0.998)	0.05	0.14	-7.2	<0.001	0.676 (-0.077-0.896)	2.42	6.72
Unenhanced - Portal-Venous phase	-3.1	<0.001	0.994 (0.778-0.999)	0.15	0.42	-1.1	<0.001	0.992 (0.743-0.998)	0.06	0.17	-7.8	<0.001	0.665 (-0.059-0.903)	1.83	5.06
Arterial phase - Portal-Venous phase	-2.2	<0.001	0.996 (0.953-0.999)	0.12	0.35	-0.7	<0.001	0.995 (0.940-0.999)	0.05	0.13	-0.6	0.161	0.949 (0.911-0.971)	0.68	1.90

Abbreviations: HU, Hounsfield Units; ICC, Intraclass Correlation Coefficient; SDC, Smallest Detectable Change; SEM, Standard Error of the Measurement

Table 2. Cohen's κ's for the assessment of low skeletal muscle mass and low skeletal muscle density using unenhanced, arterial, and portal-venous phases on CT.

		Unenhanced phase	Arterial phase
Arterial phase	Low mass	0.959	-
	Low density	0.400	
Portal-Venous phase	Low mass	0.920	0.879
	Low density	0.266	0.680

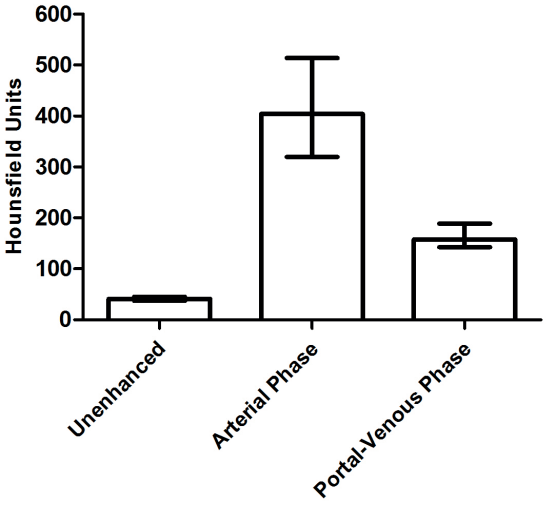


Figure 2. Median intraluminal aorta attenuation per contrast-enhancement phase. The whiskers represent the interquartile range.

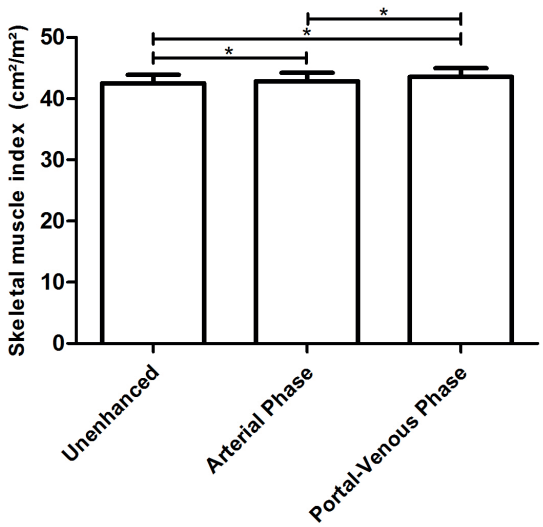
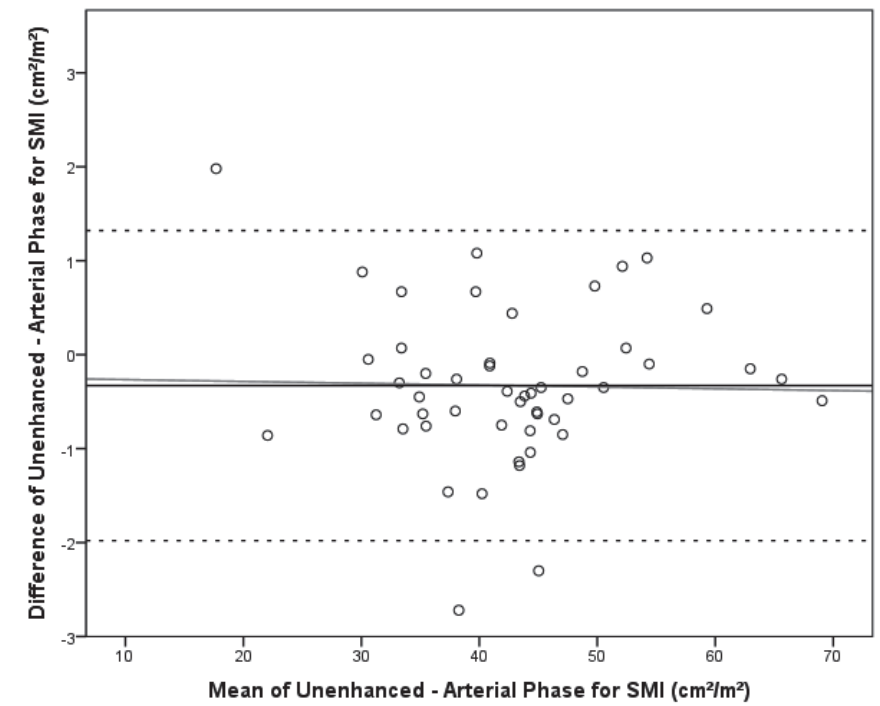
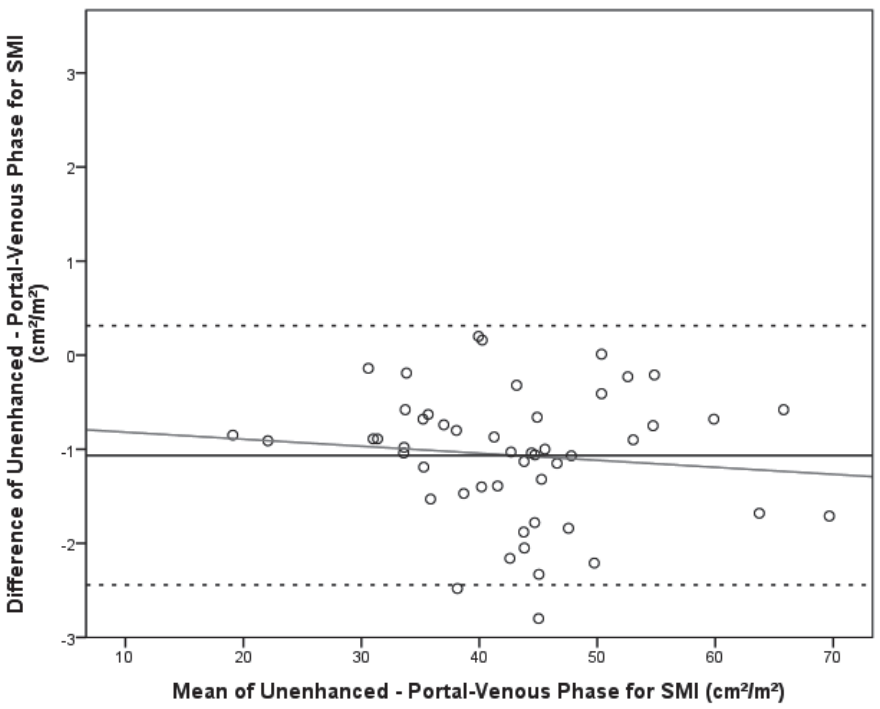


Figure 3. Mean skeletal muscle index per contrast-enhancement phase. The whiskers represent the standard error of the mean. * indicates statistically significant difference





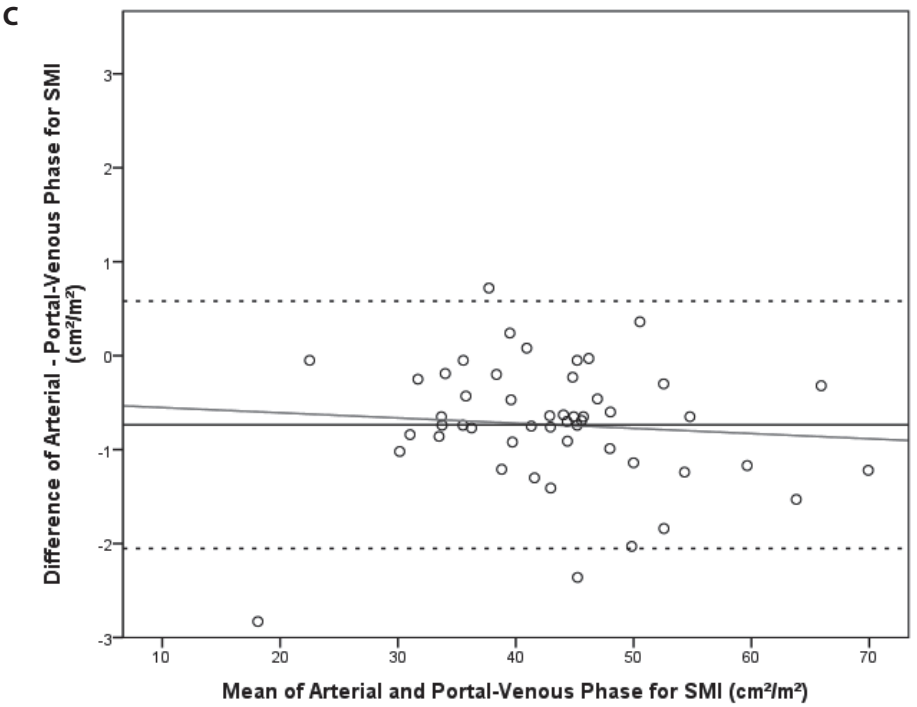


Figure 4. Bland Altman plots with 95% limits of agreement for the comparison of the cross sectional muscle area (SMI in cm^2/m^2) of the unenhanced with arterial phase (A), unenhanced with the portal-venous phase (B) and arterial with the portal-venous phase (C). The solid black line represents the mean difference, the striped lines represent the mean ± 1.96 standard deviations, and the grey line represents the regression slope.

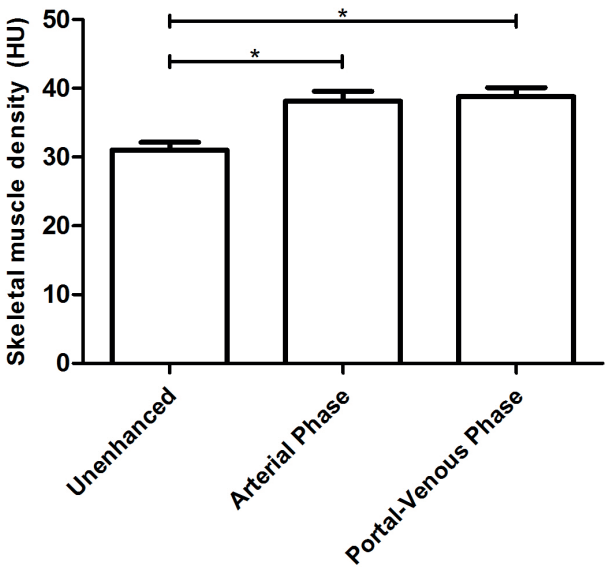
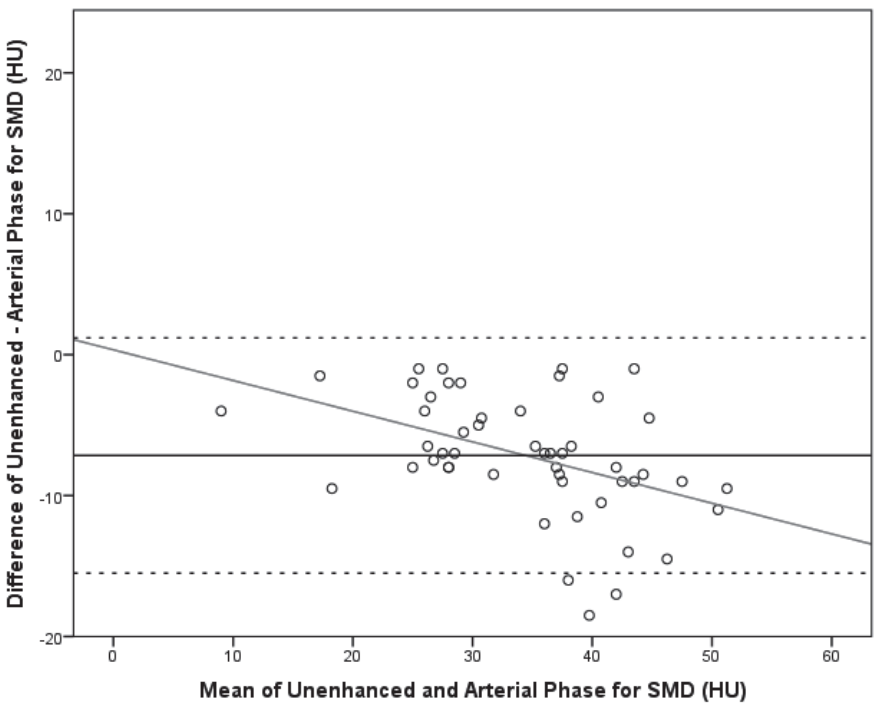


Figure 5. Mean skeletal muscle density per contrast-enhancement phase. The whiskers represent the standard error of the mean. * indicates statistically significant difference

Mean skeletal muscle density did not significantly differ between patients receiving 120 or 150 ml of contrast medium in any contrast-enhancement phase. Bland Altman plots with 95% limits of agreement for the skeletal muscle density are shown in figure 6. There was a proportional systematic bias for the comparison of the unenhanced phase with the arterial (p=0.001) and portal-venous (p=0.007) phase, but not for the comparison of the arterial with the portal-venous phase (p=0.113). The ICCs for the unenhanced phase compared with the arterial (0.676) and portal-venous (0.665) phase were considered fair to good, whereas the ICC between the arterial and portal-venous phase was considered excellent (0.949). The SDCs for skeletal muscle density measurements were considerably higher than for skeletal muscle mass measurements. The mean difference in skeletal muscle density between the arterial and venous contrast-enhancement phases (-0.6 HU) was within the SDC (1.90 HU) (table 1). According to the cut-off values defined by Martin *et al*. [24], 40 (80%) patients were considered to have low skeletal muscle density using unenhanced CT, compared with 25 (50%) patients using the arterial phase and 19 (38%) patients using the portal-venous phase. This resulted in Cohen's κ's of 0.400 (unenhanced versus arterial phase), 0.266 (unenhanced versus portal-venous phase), and 0.680 (arterial versus porta-venous phase) (table 2).

A



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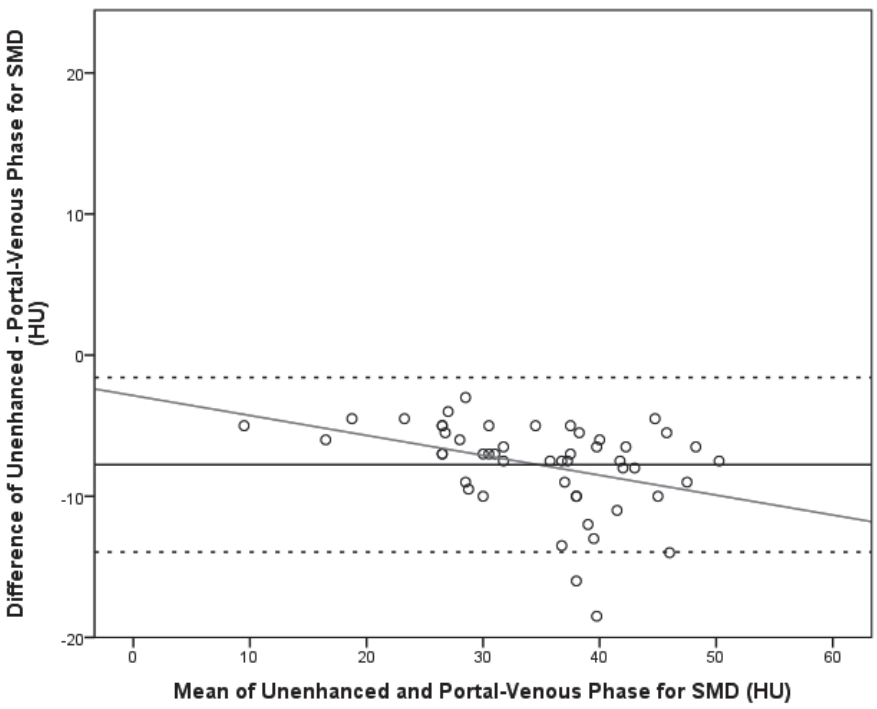

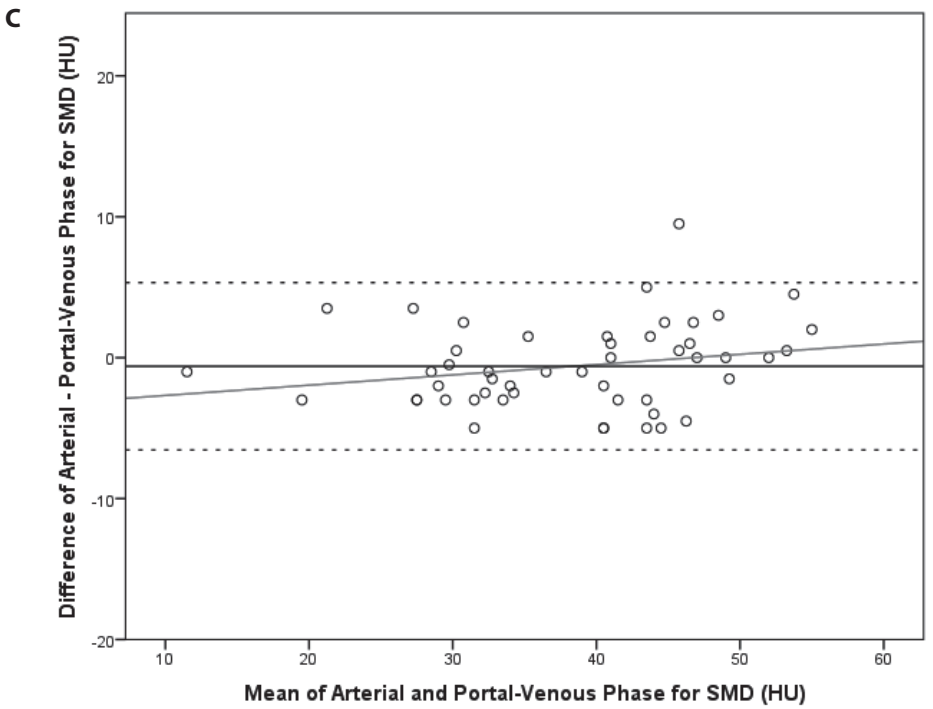

Figure 6. Bland Altman plots with 95% limits of agreement for the comparison of the mean skeletal muscle density (SMD in Hounsfield Units [HU]) of the unenhanced with arterial phase (A), unenhanced with the portal-venous phase (B) and arterial with the portal-venous phase (C). The solid black line represents the mean difference, the striped lines represent the mean ± 1.96 standard deviations, and the grey line represents the regression slope.

DISCUSSION

This is the first study to demonstrate differences in CT-based skeletal muscle mass and skeletal muscle density measurements due to different stages of contrast-enhancement in multiphase CT. Importantly, although statistically significant differences in skeletal muscle mass were found between contrast-enhancement phases, these could be considered as not clinically relevant in contrast with the differences found for skeletal muscle density measurements.

The influence of CT-assessed sarcopenia on treatment outcome has increasingly gained interest last years. Sarcopenia is associated with increased vulnerability, postoperative complications and mortality, chemotherapy toxicity, and overall survival [4, 5]. Recently, skeletal muscle density has been identified as a prognostic factor in various populations, whereas skeletal muscle mass was not [9-12, 20]. Skeletal muscle density, expressed as the mean Hounsfield unit value of the selected skeletal muscle area, is correlated with skeletal muscle lipid content [29]. Furthermore, low skeletal muscle mass is associated with increased (dose-limiting) chemotherapy toxicity [8, 30-32], and may be a superior measure to dose chemotherapy rather than body surface area which is currently being used [33].

Particularly in cancer patients, CT is considered the gold standard to measure skeletal muscle mass and density because it is routinely being performed (i.e. for diagnosis, treatment planning, and treatment evaluation) and consequently widely available [1, 15, 34]. CT-based assessment of skeletal muscle mass is an easy and reliable method correlated with total body skeletal muscle mass and known for its excellent inter- and intra-observer agreement [18, 35]. However, previous studies on the association between skeletal muscle density and treatment outcome, did not report whether unenhanced or contrast-enhanced CT images were used to measure skeletal muscle density [9-12, 20]. Results should therefore be interpreted with caution. To increase comparability within and between studies, based on our results, we recommend performing measurements in portal-venous contrast-enhanced phase CT examinations, as this phase is routinely being performed in cancer patients. Moreover, identification of various tissues is easier on contrast-enhanced CT examinations due to increased attenuation differences. When previously established cut-off values (e.g., those defined by Martin *et al.* [24]) are used, measuring skeletal muscle density on unenhanced or contrast-enhanced CT may lead to over- or underestimating the number of patients with low skeletal muscle density, respectively. This explains the poor Cohen's κ's for the classification of patients' skeletal muscle density in the current study. Therefore, we recommend to at least report the

contrast-enhancement phase used to measure skeletal muscle mass and density. Ultimately, one should seek for consensus which contrast-enhancement phase should preferably be used.

Recently, promising results to reverse cancer-induces skeletal muscle wasting have been described in animal studies [36]. Currently, multiple trials are being performed in humans to investigate drugs for the treatment of cachexia [37]. However, the general opinion is that treatment of cachexia should be multimodal, of which nutritional intervention is one modality [14, 38, 39]. Treatment strategies may be adapted as well, depending on the cancer-induced muscle loss. Body composition measures assessed on CT may guide the indication and effectiveness of these therapies.

Although all CTs used for this study have been performed within a relatively short time frame (2009-2015) in one center only, the possible use of different type of CT scanners may have led to differences in observations between patients. Indeed, there is a difference in density measurements between different vendors. However, all examinations included in this study were performed on Siemens (Erlangen, Germany) CT scanners. All scanners were calibrated daily and calibrated using a phantom monthly. Furthermore, we used identical scanning protocols for all patients, reducing differences in measurements resulting from technique variation. Nevertheless, variations on these protocols may have occurred.

Furthermore, contrast distribution depends on various factors, such as cardiac output, vascular status [40], which were unknown for the current study population, and body weight and body lean mass. However, one may expect that the influence of these factors on the uptake of contrast medium by skeletal muscles of the core are minimal in rest. Also, scanning protocols using thresholds to start scanning correct for this. Consequently, these influences may be considered negligible. After all, we did not find significant differences in mean skeletal muscle density between patients receiving 120 ml or 150 ml of contrast medium. Furthermore, each patient forms its reference in this study as paired t-tests have been used. Furthermore, the variation in aortic intraluminal attenuation measurements in each contrast phase was relatively small. Contrary to a previous study [18], consecutive measures in the various contrast-enhancement phases were not performed on identical slices, since patients' movements and differences in breath-hold may have led to variations in the level on which measurements were performed. These factors could, however, better be controlled for in a future prospective study. Moreover, although single-slice cross-sectional areas are strongly correlated with whole body skeletal muscle mass, this remains an estimation only, which may introduce a potential error of several kilograms. Finally, inter-observer variation may have led to

measurement differences, although the inter-observer agreement for skeletal muscle mass measurements is excellent [18] and the mean of the two observers was used for analyses to further minimize inter-observer differences.

In conclusion, significant and clinically relevant differences in skeletal muscle density were observed between contrast-enhancement phases, whereas significant but not clinically relevant differences were found in skeletal muscle mass measurements. We recommend using the portal-venous phase of contrast-enhanced CT for studies that describe the association between skeletal muscle density and outcome measures to improve comparability of studies.

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REFERENCES

1. Fearon K, Strasser F, Anker SD, Bosaeus I, Bruera E, Fainsinger RL, et al. Definition and classification of cancer cachexia: an international consensus. Lancet Oncol 2011; 12: 489-95.
2. Rolland Y, Abellan van Kan G, Gillette-Guyonnet S, Vellas B. Cachexia versus sarcopenia. Current opinion in clinical nutrition and metabolic care 2011; 14: 15-21.
3. Lieffers JR, Mourtzakis M, Hall KD, McCargar LJ, Prado CM, Baracos VE. A viscerally driven cachexia syndrome in patients with advanced colorectal cancer: contributions of organ and tumor mass to whole-body energy demands. The American journal of clinical nutrition 2009; 89: 1173-9.
4. Levolger S, van Vugt JL, de Bruin RW, IJzermans JN. Systematic review of sarcopenia in patients operated on for gastrointestinal and hepatopancreatobiliary malignancies. Br J Surg 2015; 102: 1448-58.
5. van Vugt JL, Levolger S, de Bruin RW, van Rosmalen J, Metselaar HJ, IJzermans JN. Systematic Review and Meta-Analysis of the Impact of Computed Tomography-Assessed Skeletal Muscle Mass on Outcome in Patients Awaiting or Undergoing Liver Transplantation. Am J Transplant 2016; 16: 2277-92.
6. Joglekar S, Nau PN, Mezhir JJ. The impact of sarcopenia on survival and complications in surgical oncology: A review of the current literature. J Surg Oncol 2015; 112: 503-9.
7. Gani F, Buettner S, Margonis GA, Sasaki K, Wagner D, Kim Y, et al. Sarcopenia predicts costs among patients undergoing major abdominal operations. Surgery 2016.
8. Anandavadivelan P, Brismar TB, Nilsson M, Johar AM, Martin L. Sarcopenic obesity: A probable risk factor for dose limiting toxicity during neo-adjuvant chemotherapy in esophageal cancer patients. Clin Nutr 2016; 35: 724-30.
9. Hayashi N, Ando Y, Gyawali B, Shimokata T, Maeda O, Fukaya M, et al. Low skeletal muscle density is associated with poor survival in patients who receive chemotherapy for metastatic gastric cancer. Oncol Rep 2016; 35: 1727-31.
10. Sabel MS, Lee J, Cai S, Englesbe MJ, Holcombe S, Wang S. Sarcopenia as a prognostic factor among patients with stage III melanoma. Ann Surg Oncol 2011; 18: 3579-85.
11. Chu MP, Lieffers J, Ghosh S, Belch AR, Chua NS, Fontaine A, et al. Skeletal muscle radio-density is an independent predictor of response and outcomes in follicular lymphoma treated with chemoimmunotherapy. PLoS One 2015; 10: e0127589.
12. Antoun S, Lanoy E, Iacovelli R, Albiges-Sauvin L, Loriot Y, Merad-Taoufik M, et al. Skeletal muscle density predicts prognosis in patients with metastatic renal cell carcinoma treated with targeted therapies. Cancer 2013; 119: 3377-84.
13. Rollins KE, Tewari N, Ackner A, Awwad A, Madhusudan S, Macdonald IA, et al. The impact of sarcopenia and myosteatosis on outcomes of unresectable pancreatic cancer or distal cholangiocarcinoma. Clin Nutr 2016; 35: 1103-9.

14. Fearon KC. The 2011 ESPEN Arvid Wretlind lecture: cancer cachexia: the potential impact of translational research on patient-focused outcomes. Clin Nutr 2012; 31: 577-82.

15. Prado CM, Birdsell LA, Baracos VE. The emerging role of computerized tomography in assessing cancer cachexia. Curr Opin Support Palliat Care 2009; 3: 269-75.

16. Gibson DJ, Burden ST, Strauss BJ, Todd C, Lal S. The role of computed tomography in evaluating body composition and the influence of reduced muscle mass on clinical outcome in abdominal malignancy: a systematic review. Eur J Clin Nutr 2015; 69: 1079-86.

17. Prado CM. Body composition in chemotherapy: the promising role of CT scans. Current opinion in clinical nutrition and metabolic care 2013; 16: 525-33.

18. van Vugt JL, Levolger S, Gharbharan A, Koek M, Niessen WJ, Burger JW, et al. A comparative study of software programs for cross-sectional skeletal muscle and adipose tissue measurements on abdominal computed tomography scans of rectal cancer patients. J Cachexia Sarcopenia Muscle 2016: *In press*.

19. Kormano MJ, Goske MJ, Hamlin DJ. Attenuation and contrast enhancement of gynecologic organs and tumors in CT. Eur J Radiol 1981; 1: 307-11.

20. Van Rijssen LB, van Huijgevoort NC, Coelen RJ, Tol JA, Haverkort EB, Nio CY, et al. Skeletal Muscle Quality is Associated with Worse Survival After Pancreatoduodenectomy for Periampullary, Nonpancreatic Cancer. Ann Surg Oncol 2016.

21. Ogden CL, Carroll MD, Kit BK, Flegal KM. Prevalence of obesity and trends in body mass index among US children and adolescents, 1999-2010. Jama 2012; 307: 483-90.

22. van Vledder MG, Levolger S, Ayez N, Verhoef C, Tran TC, IJzermans JN. Body composition and outcome in patients undergoing resection of colorectal liver metastases. Br J Surg 2012; 99: 550-7.

23. van Vugt JL, Levolger S, Gharbharan A, Koek M, Niessen WJ, Burger JW, et al. A comparative study of software programmes for cross-sectional skeletal muscle and adipose tissue measurements on abdominal computed tomography scans of rectal cancer patients. J Cachexia Sarcopenia Muscle 2017; 8: 285-97.

24. Martin L, Birdsell L, Macdonald N, Reiman T, Clandinin MT, McCargar LJ, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol 2013; 31: 1539-47.

25. Bland JM, Altman DG. Statistical methods for assessing agreement between two methods of clinical measurement. Lancet 1986; 1: 307-10.

26. Ludbrook J. Statistical techniques for comparing measurers and methods of measurement: a critical review. Clin Exp Pharmacol Physiol 2002; 29: 527-36.

27. Bruynesteyn K, Boers M, Kostense P, van der Linden S, van der Heijde D. Deciding on progression of joint damage in paired films of individual patients: smallest detectable difference or change. Ann Rheum Dis 2005; 64: 179-82.

28. Shrout PE, Fleiss JL. Intraclass correlations: uses in assessing rater reliability. Psychol Bull 1979; 86: 420-8.

29. Goodpaster BH, Kelley DE, Thaete FL, He J, Ross R. Skeletal muscle attenuation determined by computed tomography is associated with skeletal muscle lipid content. J Appl Physiol (1985) 2000; 89: 104-10.

30. Prado CM, Baracos VE, McCargar LJ, Mourtzakis M, Mulder KE, Reiman T, et al. Body composition as an independent determinant of 5-fluorouracil-based chemotherapy toxicity. Clin Cancer Res 2007; 13: 3264-8.

31. Baracos VE, Reiman T, Mourtzakis M, Gioulbasanis I, Antoun S. Body composition in patients with non-small cell lung cancer: a contemporary view of cancer cachexia with the use of computed tomography image analysis. The American journal of clinical nutrition 2010; 91: 1133S-7S.

32. Ali R, Baracos VE, Sawyer MB, Bianchi L, Roberts S, Assenat E, et al. Lean body mass as an independent determinant of dose-limiting toxicity and neuropathy in patients with colon cancer treated with FOLFOX regimens. Cancer Med 2016; 5: 607-16.

33. Crosby V, D'Souza C, Bristow C, Proffitt A, Hussain A, Potter V, et al. Can body composition be used to optimize the dose of platinum chemotherapy in lung cancer? A feasibility study. Support Care Cancer 2016.

34. Yip C, Dinkel C, Mahajan A, Siddique M, Cook GJ, Goh V. Imaging body composition in cancer patients: visceral obesity, sarcopenia and sarcopenic obesity may impact on clinical outcome. Insights Imaging 2015; 6: 489-97.

35. Shen W, Punyanitya M, Wang Z, Gallagher D, St-Onge MP, Albu J, et al. Total body skeletal muscle and adipose tissue volumes: estimation from a single abdominal cross-sectional image. J Appl Physiol (1985) 2004; 97: 2333-8.

36. Zhou X, Wang JL, Lu J, Song Y, Kwak KS, Jiao Q, et al. Reversal of cancer cachexia and muscle wasting by ActRIIB antagonism leads to prolonged survival. Cell 2010; 142: 531-43.

37. Dingemans AM, de Vos-Geelen J, Langen R, Schols AM. Phase II drugs that are currently in development for the treatment of cachexia. Expert Opin Investig Drugs 2014; 23: 1655-69.

38. Gullett NP, Mazurak VC, Hebbar G, Ziegler TR. Nutritional interventions for cancer-induced cachexia. Curr Probl Cancer 2011; 35: 58-90.

39. Baldwin C. The effectiveness of nutritional interventions in malnutrition and cachexia. Proc Nutr Soc 2015; 74: 397-404.

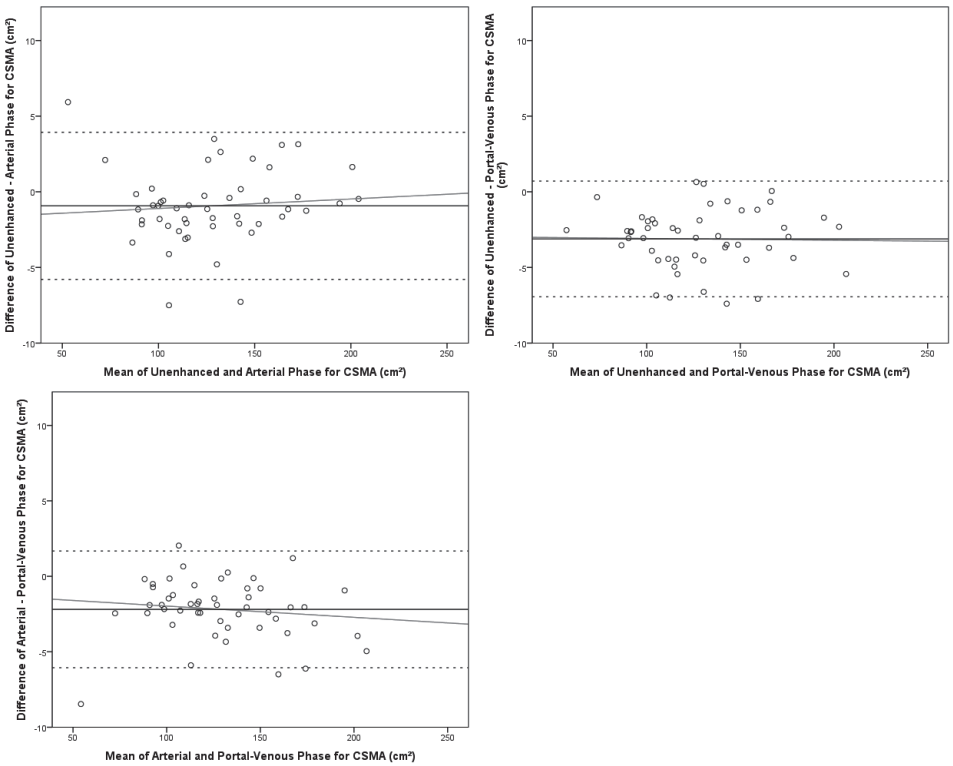
40. Fleischmann D, Kamaya A. Optimal vascular and parenchymal contrast enhancement: the current state of the art. Radiol Clin North Am 2009; 47: 13-26.

SUPPLEMENTARY MATERIAL

Supplementary table 1. Baseline characteristics of the included patients.

Characteristic	**N = 50**
Age, years	62 (50-69)
Indication CT	
Pancreatic cancer	16 (32.0)
Perihilar cholangiocarcinoma	5 (10.0)
Hepatocecullar carcinoma	21 (42.0)
Liver transplantation evaluation	8 (16.0)
BMI, kg/m^2	23.9 (21.0-26.5)
< 20	5 (10.0)
20-24.9	26 (52.0)
25-29.9	15 (30.0)
≥ 30	4 (8.0)

Continuous parameters are shown as median with interquartile range, dichotomous parameters as count with percentages.
Abbreviations: CT, computed tomography; BMI, body mass index.



Supplementary figure 1. Bland Altman plots with 95% limits of agreement for the comparison of skeletal muscle index (CSMA in cm^2/m^2) of the unenhanced with arterial phase (A), unenhanced with the portal-venous phase (B) and arterial with the portal-venous phase (C). The solid black line represents the mean difference, the striped lines represent the mean ± 1.96 standard deviations, and the grey line represents the regression slope.

CHAPTER 4

Estimated Skeletal Muscle Mass and Density Values Measured on Computed Tomography Examinations Established in over One Thousand Healthy Subjects

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Submitted

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ABSTRACT

Background: There is still debate regarding adequate cut-off points to categorize patients as having sarcopenia (low skeletal muscle mass) based on computed tomography (CT) measurements. No international consensus has been reached yet. Moreover, there is insufficient knowledge on skeletal muscle mass in healthy persons, particularly in a Western-European population.

Methods: Skeletal muscle mass at the level of the third lumbar vertebra (skeletal muscle index, cm^2/m^2) and density (Hounsfield units, HU) were measured on contrast-enhanced CT images in live kidney donors, which may be considered as healthy subjects. Differences between sex, body mass index (BMI), age groups, and ASA classification were assessed. Groups were compared using the Mann-Whitney-U and Kruskal-Wallis tests.

Results: Of the 1073 included patients, 499 (46.5%) were male. Median age was 51 years and median BMI 25.4 kg/m^2. Male gender, increased age, and increased BMI were significantly associated with both an incremental skeletal muscle mass and density. Nomograms including these parameters were developed to calculate the estimated skeletal muscle mass and density of a healthy subject.

Conclusions: Skeletal muscle density and mass were significantly associated with sex, age, and BMI in a large cohort of Western-European healthy subjects. The newly developed nomograms may be used to calculate the estimated healthy skeletal muscle mass for individuals in patient populations.

INTRODUCTION

To perform body composition analyses, a precise method which is applicable with high specificity on a large scale is a precondition. In 1979, Heymsfield *et al.* reported the use of computed tomography (CT) to measure skeletal muscle and visceral adipose tissue mass [1]. Kvist *et al.* were the first to assess whole-body adipose tissue volumes with computed tomography studies using just several slices in 1986 [2]. A standardized part of the body with images at specific skeletal landmarks were chosen [3]. Shen *et al.* showed high correlations between two-dimensional abdominal skeletal muscle and adipose tissue areas measured on just a single slice and their respective three-dimensional total body volumes, after examination in a large sample of diverse subjects in 2004 [4].

In most later studies the third lumbar vertebra (L3) was usually chosen as the level to perform these measurements. Prado *et al.* were first to show that measurements performed at the level of L3 were related to impaired outcomes in malignancies of the upper respiratory and digestive tract in 2008 [5]. Many publications followed in various populations [6, 7]. This CT-based method was also shown to provide more detail than the formerly used dual X-ray absorptiometry (DXA) and bioelectrical impedance analysis (BIA) [8].

There is still debate regarding adequate cut-off points to categorize patients as having sarcopenia. Prado *et al.* used cut-off values based on risk stratification [5]. These have been used commonly, yet some studies use different cut-off values [9-11]. No international consensus has been reached yet, and different cut-off values for skeletal muscle mass lead to variations in the reported prevalence of sarcopenia [12]. Moreover, there is insufficient knowledge on skeletal muscle mass in healthy persons, particularly in Western-European populations. Therefore, the aim of this study was to define references values for skeletal muscle mass and density measurements on abdominal CT examinations established in healthy subjects.

METHODS

Subjects

All subjects ≥18 years of age participating in a live kidney donation program between May 2010 and August 2015 in Erasmus MC University Medical Center (Rotterdam, the Netherlands) and between January 2003 and January 2012 in Radboud University Medical Center (Nijmegen, the Netherlands) were identified. Sex, age at the moment of the abdominal CT examination, and self-reported body height and body length were recorded. The body mass index (BMI, kg/m^2) was calculated. Furthermore, the American Society of Anesthesiologists (ASA) classification was used to categorize physical status. This study was approved by the Institutional Review Boards (IRB) of both centers and a waiver for informed consent was granted.

Skeletal muscle mass and density measurements

Routinely performed contrast-enhanced abdominal CT examinations to evaluate vascular anatomy of the kidney donors were collected. The skeletal muscle mass and density measurements were performed as previously described [13]. In short, the cross-sectional skeletal muscle area (cm^2) was quantified at the level of the third lumbar vertebra (L3) using Hounsfield Units (HU) thresholds (i.e. -30 HU to +150 HU) by manually outlining the following muscle groups: the psoas, paraspinal, and abdominal wall muscles. The cross-sectional skeletal muscle area (CSMA) was adjusted for subjects' body height, resulting in the skeletal muscle index (SMI, cm^2/m^2). The mean muscle density (in HU), a measure for intramuscular adipose tissue infiltration, of the selected skeletal muscle tissue was also recorded. Subjects with low skeletal muscle mass and density were classified according to cut-off values that are currently most used [11]. Only patients with contrast-enhanced CT examinations were included, since significant differences have been reported in outcome measure between contrast-enhanced and unenhanced CT images [14].

Statistical analyses

Categorical data are presented as counts with percentages, whereas continuous data are presented as mean (± standard deviation [SD]) or median (interquartile range [IQR]) depending on the normality of data distribution. Groups were compared using Student's t-tests or one-way ANOVA analysis in the case of normal distribution and the Mann-Whitney U or Kruskal-Wallis H test in the case of not normally distributed data. After linearity was assessed, a multivariable linear regression analysis was used to investigate the association between skeletal muscle density and mass and various patient characteristics. The linear regression coefficients were used to create nomograms. Two-

sided p-values <0.05 were considered statistically significant. Analyses were performed using SPSS for Windows version 22 (IBM Corp., Armonk, NY, USA) and the RMS package in R version 3.03 (http://www.r-project.org).

RESULTS

Subjects

In total, 1163 subjects were identified, of whom a CT examination was available in 1145 patients. In 56 subjects not all skeletal muscle mass of interested was depicted on CT, 10 subjects only had an unenhanced CT, 3 had a CT without the level of L3 depicted, 2 had CTs with artefacts that significantly influenced measurements, and in 1 subject we experienced technical difficulties that did not allow measurements. The remaining 1073 subjects (92.3%) formed the study cohort. Of these patients, the median age was 51 (IQR 41-59) years and 499 (46.5%) were male. All baseline characteristics are shown in table 1.

Table 1. Baseline characteristics.

Characteristic	N = 1073
Sex	
Men	499 (46.5)
Women	574 (53.5)
Age, years	51 (41-59)
Body height, m	1.72 (1.66-1.79)
Men	1.79 (1.74-1.84)
Women	1.67 (1.62-1.71)
Body weight, kg	76 (67-86)
Men	83 (75-91)
Women	70 (63-78)
BMI, kg/m^2	25.4 (23.4-28.4)
Men	25.7 (23.9-28.5)
Women	24.9 (23.0-28.1)
ASA classification	
1	686 (63.9)
2	374 (34.9)
3	4 (0.4)
Missing	9 (0.8)

Abbreviations: ASA, American Society of Anesthesiologists.

The median CSMA for all patients was 137.6 (IQR 117.7-172.5) cm^2, corresponding to a median SMI of 47.4 (IQR 41.9-54.6) cm/m^2. CSMA was significantly higher in men (174.2 [IQR 159.0-193.0) cm^2) than in women (119.2 [IQR 109.6-130.3]), p<0.001. The median muscle attenuation for all patients was 44 (IQR 38-49) HU.

Influence of sex, age, BMI, and ASA classification on skeletal muscle mass and density

The association between sex, age, BMI, and ASA classification with skeletal muscle index and skeletal muscle attenuation is shown in table 2. Both skeletal muscle mass and density were significantly higher in males compared with females (figure 1) and significantly decreased with and incremental increase in age (figure 2). Skeletal muscle mass significantly increased with an incremental increase in BMI, whereas skeletal muscle attenuation showed a significant decrease (figure 3). No significant differences were found between patients with ASA classification 1 versus patients with ASA classification 2-3 with regards to skeletal muscle mass, whereas patients with ASA classification 1 had a significantly higher skeletal muscle density compared with patients with ASA classification 2-3 (figure 4).

Classification of subjects using established cut-off values

According to the cut-off values by Martin *et al.* [11], a total of 113 (10.5%) patients were considered to have low skeletal muscle mass and 309 (28.8%) patients to have low skeletal muscle density. The prevalence of low skeletal muscle mass significantly differed between age groups (13.3% in patients aged 18-39, 54.4% in patients aged 40-59, and 32.4% in patients aged ≥60, $p<0.001$). Also, the prevalence of low skeletal muscle density significantly differed between age groups (5.3% in patients aged 18-39, 47.8% in patients aged 40-59, and 46.9% in patients aged ≥60, $p<0.001$). Differences between patients with low and high skeletal muscle mass, and low and high skeletal muscle density, are summarized in table 3.

Multivariable linear regression analysis for factors associated with skeletal muscle mass and density and the development of nomograms

A multivariable linear regression analysis showed that sex, age, and BMI were independently associated with both skeletal muscle mass and density, whereas ASA classification was not (table 4).

Using these parameters, nomograms were created for skeletal muscle index (figure 5) and skeletal muscle density (figure 6) stratified for sex. For example, a 40-year-old man (22 points) with a BMI of 28 (42 points) has a total of 64 points, corresponding to a SMI of 56 cm^2/m^2 (figure 5a). Another example: a 65-year-old woman (25 points) with a BMI of 20 (18 points) has a total of 43 points, corresponding with a muscle density of 30 HU (figure 6b). Finally, an online calculator was developed (Supplementary material).

Table 2. The association between sex, age, body mass index (BMI), and American Society for Anesthesiologists (ASA) classification and skeletal muscle mass and density measures.

Characteristic	N (%)	Skeletal muscle index, cm^2/m^2	p-value	Skeletal muscle attenuation, HU	p-value
Sex			<0.001		<0.001
Men	499 (46.5)	54.4 (49.7-60.2)		45 (40-50)	
Women	574 (53.5)	42.8 (39.4-46.9)		43 (36-49)	
Age, years					
Men			<0.001		<0.001
18-39	119 (23.8)	58.5 (52.9-63.5)		50 (45-55)	
40-59	268 (53.7)	54.3 (49.6-59.7)		44 (40-48)	
≥60	112 (22.4)	52.4 (47.6-56.0)		41 (36-45)	
Women			0.005		<0.001
18-39	112 (19.5)	44.0 (40.2-48.0)		49 (44-54)	
40-59	324 (56.4)	43.1 (39.5-46.9)		43 (37-48)	
≥60	138 (24.0)	41.3 (38.5-46.1)		37 (32-42)	
BMI, kg/m^2					
Men			<0.001		<0.001
<20	19 (3.8)	46.4 (44.3-49.7)		48 (44-54)	
20.0-24.9	185 (37.1)	51.2 (46.7-55.3)		46 (42-52)	
25.0-29.9	226 (45.3)	57.2 (52.4-61.2)		43 (38-48)	
≥30	69 (13.8)	60.3 (55.0-65.2)		42 (34-47)	
Women			<0.001		<0.001
<20	33 (5.7)	37.7 (36.4-41.9)		51 (45-55)	
20.0-24.9	256 (44.6)	41.5 (38.5-45.1)		44 (40-50)	
25.0-29.9	211 (36.8)	44.1 (40.0-47.6)		40 (36-46)	
≥30	74 (12.9)	47.2 (43.3-51.2)		39 (31-44)	
ASA classification*					
Men			0.172		<0.001
1	318 (64.4)	55.3 (49.8-60.4)		45 (41-51)	
2-3	176 (35.6)	53.1 (49.3-59.2)		42 (37-47)	
Women			0.133		<0.001
1	368 (64.6)	42.6 (39.3-46.5)		44 (39-50)	
2-3	202 (35.4)	43.4 (39.4-47.3)		40 (34-46)	

* 9 missing values.

Table 3. Differences between patients who have been classified as having low/high skeletal muscle mass and low/high skeletal muscle density according to Martin *et al.*.

	Low skeletal muscle mass	High skeletal muscle mass	p-value	Low skeletal muscle density	High skeletal muscle density	p-value
Sex						
Men	100 (32.4)	399 (52.2)	<0.001	34 (30.1)	465 (48.4)	<0.001
Age, years	55 (46-62)	50 (40-58)	<0.001	58 (53-65)	50 (40-58)	<0.001
BMI, kg/m^2	24.8 (22.3-27.3)	25.6 (23.7-28.7)	<0.001	28.4 (25.7-30.7)	25.0 (23.1-28.0)	<0.001
ASA classification*						<0.001
1	189 (61.6)	497 (65.7)		50 (44.2)	636 (66.9)	
2-3	118 (38.4)	260 (34.3)	0.207	63 (55.8)	315 (33.1)	

* 9 missing values.

Table 4. Multivariable linear regression analysis to identify parameters that are independently associated with skeletal muscle mass (upper analysis) and density (lower analysis).

		B	SE	98% CI	p-value
Skeletal muscle mass	Sex (female)	-11.07	0.38	-11.83; -10.32	<0.001
	Age (years)	-0.11	0.02	-0.14; -0.08	<0.001
	BMI (categories)	3.77	0.25	3.28; 4.27	<0.001
	ASA (3 vs 1-2)	-0.25	0.42	-1.08; 0.58	0.549
Skeletal muscle density	Sex (female)	-1.70	0.49	-2.65; -0.75	<0.001
	Age (years)	-0.32	0.02	-0.36; -0.28	<0.001
	BMI (categories)	-3.28	0.32	-3.90; -2.65	<0.001
	ASA (3 vs 1-2)	-0.99	0.54	-2.04; 0.07	0.066

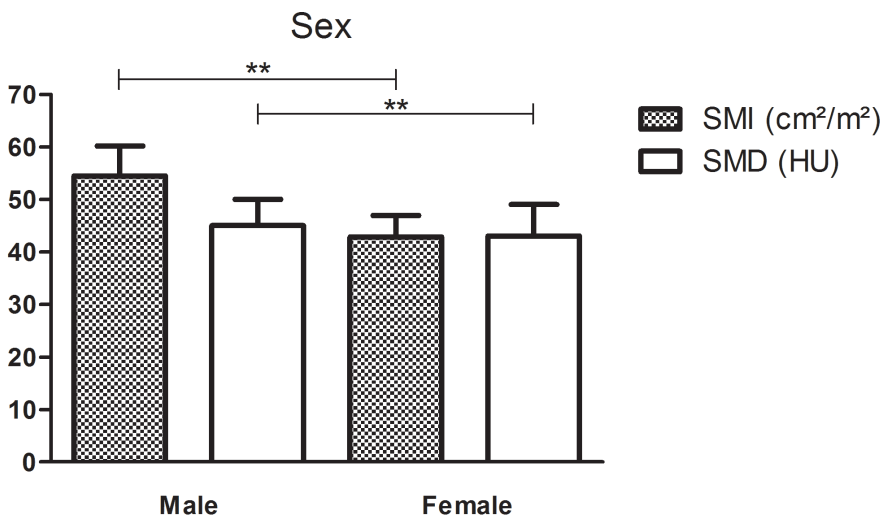

Figure 1. Skeletal muscle index (SMI) and skeletal muscle density (SMD) stratified by sex.

** indicates p<0.001.

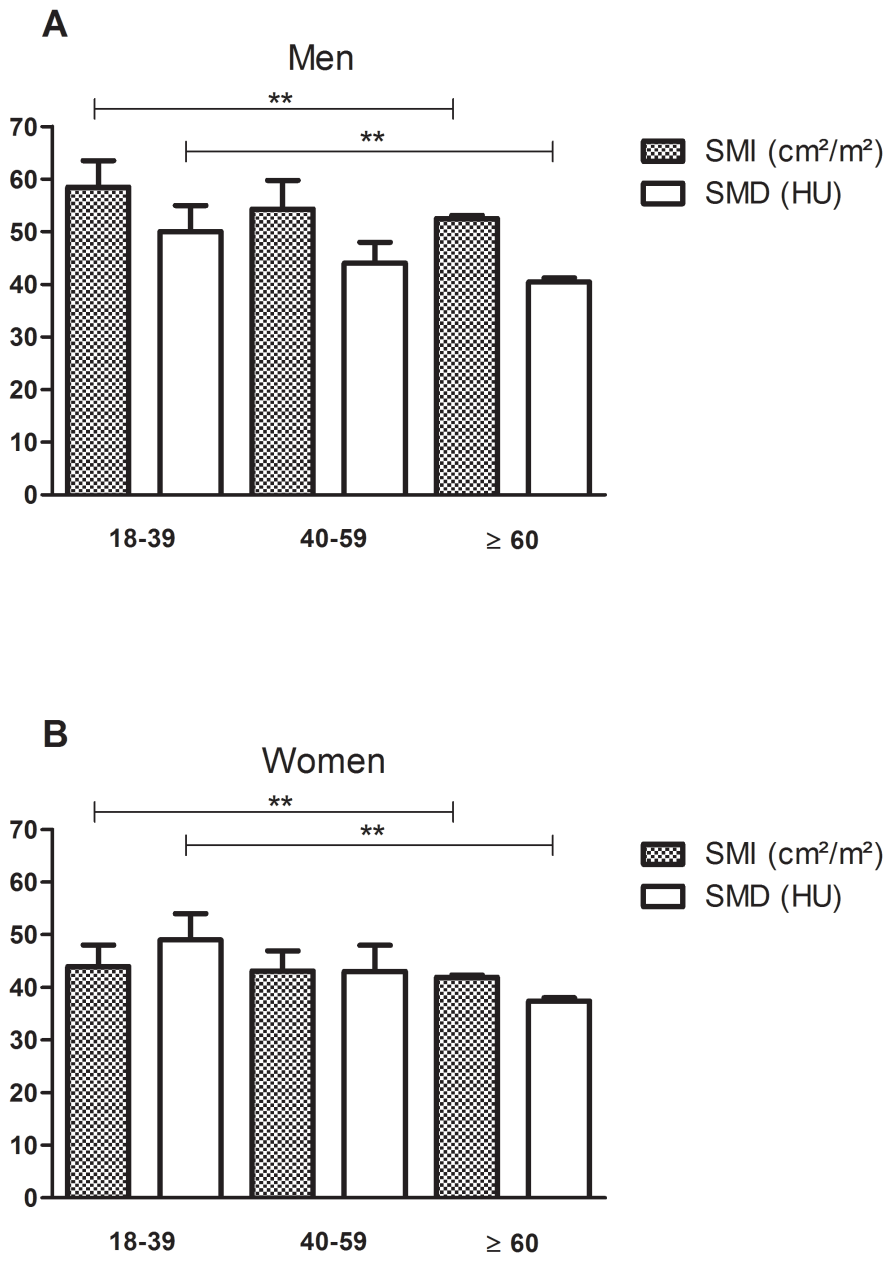

Figure 2. Skeletal muscle index (SMI) and skeletal muscle density (SMD) stratified by age groups (i.e. 18-39, 40-59, ≥60 years) for men (A) and women (B).

** indicates p<0.001.

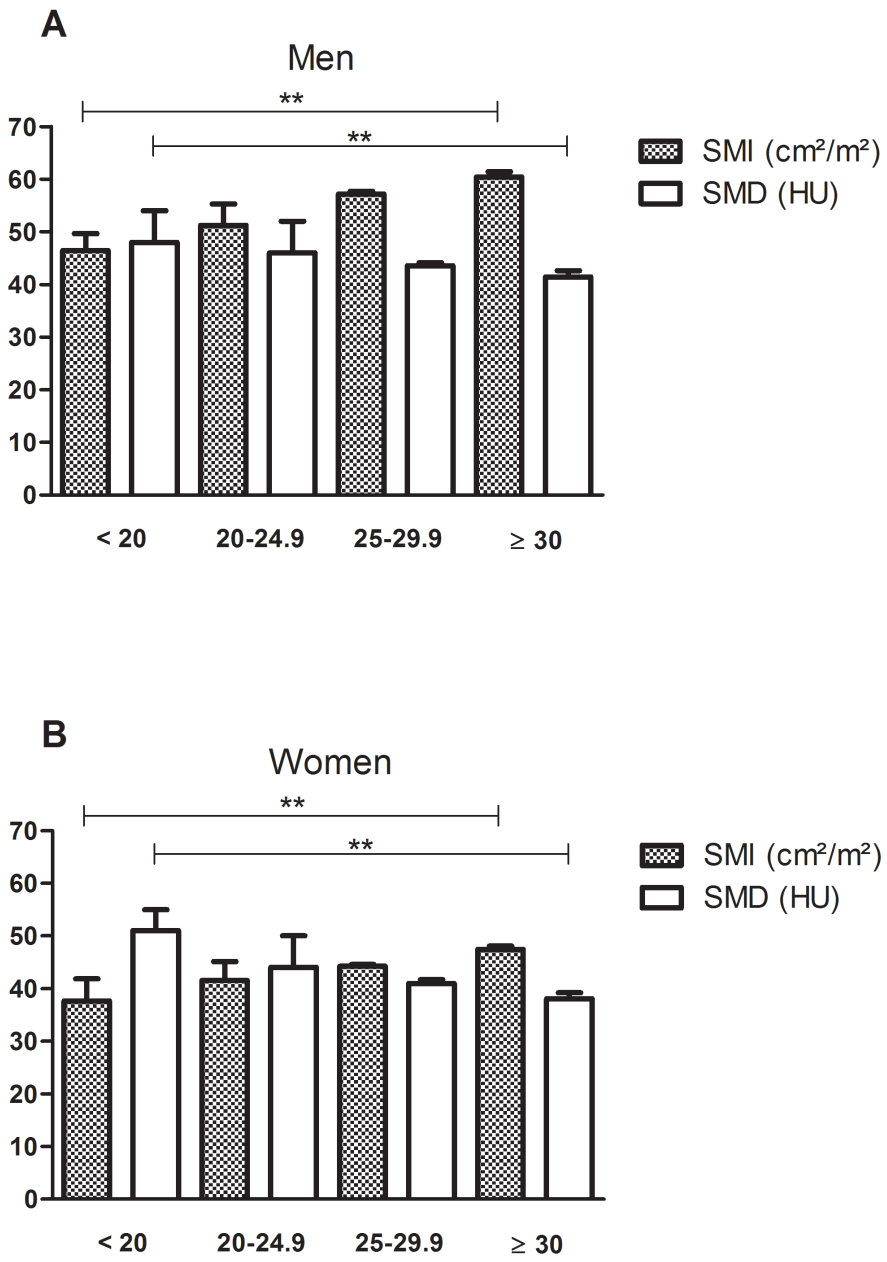

Figure 3. Skeletal muscle index (SMI) and skeletal muscle density (SMD) stratified by body mass index (BMI) groups (i.e. <20, 20-24.9, 25.0-29.0, ≥30 kg/m^2) for men (A) and women (B).

** indicates p<0.001.

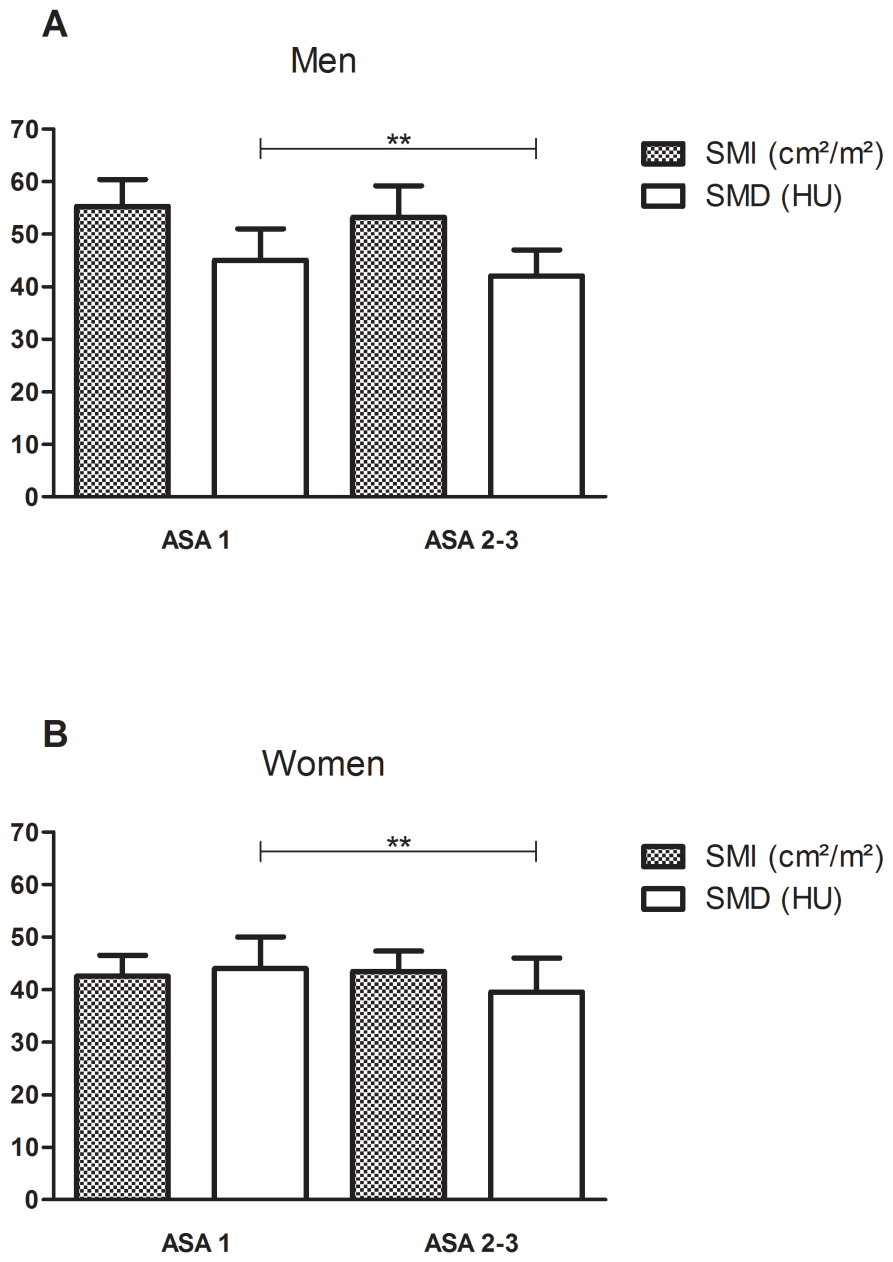

Figure 4. Skeletal muscle index (SMI) and skeletal muscle density (SMD) stratified by American Society of Anesthesiologists (ASA) classification for men (A) and women (B).

** indicates p<0.001.

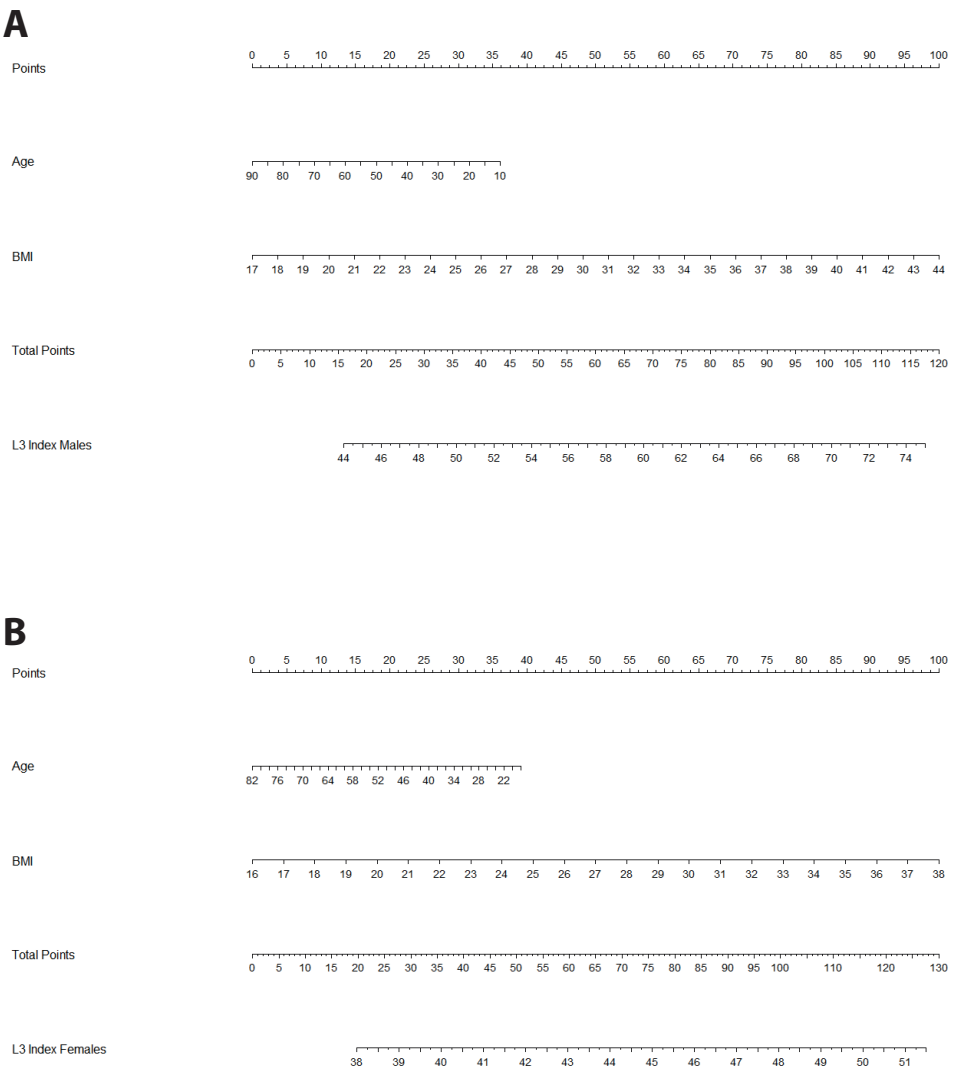

Figure 5. Nomogram showing reference values for skeletal muscle index for men (A) and women (B), adjusted for age and body mass index (BMI).

Figure 6. Nomogram showing reference values for skeletal muscle density for men (A) and women (B), adjusted for age and body mass index (BMI).

DISCUSSION

This is the first study that described skeletal muscle mass and density measurements on CT in a large population of healthy subjects. Currently, most studies classify patients as having low skeletal muscle mass or density based on cut-off values that have been established in patient specific populations, such as cancer [5, 9, 11] or liver transplantation patients [15, 16], to predict overall survival. Consequently, the reported prevalence of low CT-assessed skeletal muscle mass and density may greatly differ between studies [6, 7]. We proposed reference values, stratified for sex and adjusted for age and BMI, which may be used to classify patients as having low skeletal muscle mass. Use of these reference values may increase comparability of studies with varying populations.

Live kidney donors form a unique cohort, as they may be considered as healthy subjects rather than patients, with an equal distribution of men and women and a relatively broad range in age. A previous study among 45 healthy Japanese adults willing to donate a part of their liver also defined normal values for skeletal muscle mass measured on CT [17]. In this study, median CSMA was 155.0 cm^2 in men and 111.7 cm^2 women, which is considerably lower compared with our study. Furthermore, the incidence of overweight and obesity is lower in Asia compared with Europe and North-America [18]. This underlines the need for reference values which are specific per geographical region (i.e. European, Asian, and North-American) or ethnicity, as genetics are an important determinant of muscularity [19, 20]. Another Japanese study proposed reference values for low skeletal muscle mass based on CT in 541 adult donors for living donor liver transplantation [21]. However, skeletal muscle mass was assessed on psoas muscle measurements only rather than CSMA and skeletal muscle density was not assessed [21]. Although comparable findings were reported [21], the psoas muscle alone may not be a valid representation of total body skeletal muscle mass [22]. Lower skeletal muscle mass in younger and female patients was also observed among the general population in the United Kingdom using bioelectrical impedance analysis (BIA) [23] and among Mexican elderly using dual-energy X-ray absorptiometry (DXA) [24].

Besides sex, age, and BMI, another significant predictor for skeletal muscle mass and density is comorbidity. However, our study cohort consisted of live kidney donors, which may be considered healthy subjects. Consequently, 99.6% of our cohort had a low ASA classification (i.e. ASA 1 or 2). Therefore, we did not find an association between ASA classification and skeletal muscle mass or density. The healthy status of our subjects may also explain the relatively low prevalence of low skeletal muscle mass and density, particularly in the older patients (i.e. ≥60 years), because all patients were considered fit enough to donate one kidney.

Interestingly, a recent study showed that grip strength depends on socioeconomic status (measured using household income) [25]. Another study described the association between household income and frailty in middle-aged and older adults in Switzerland [26]. We, however, did decide not to include socioeconomic status as a parameter to defined reference values for skeletal muscle mass and density, because it would reduce its clinical applicability. Nevertheless, it would be interesting to investigate the association between body composition and socioeconomic status in a future study. Previous studies also described differences in grip strength [25] and muscle mass [24] between races. Unfortunately, we were not able to include race as a parameter, since the number of non-Caucasian patients would be too small.

Inherent to the study's retrospective character, we were not able to investigate normal values of muscle strength and gait speed, which are considered essential to diagnose sarcopenia (i.e. the involuntary age-related loss of skeletal muscle mass and function) [27]. Future prospective studies may therefore combine our reference values with measures of muscle strength and function (e.g., gait speed). Nevertheless, the association between CT-assessed skeletal muscle mass and density has frequently been described and seems a strong predictive factor in various patient populations, such as cancer patients [6] and liver transplant candidates [7]. Particularly automatic segmentation, which is currently being developed [28], may greatly increase implementation in daily practice. Another limitation of the current study is that most body weight and height measures were self-reported. This may have led to discrepancies between the true BMI and self-reported BMI. In particular, overweight individuals tend to under-report and underweight individuals to over-report [29, 30]. Although our results enable calculating the estimated normal value for skeletal muscle mass and density in patients, our findings should externally being validated in patients cohorts.

Computed tomography is routinely made as part of the diagnostic tract, preoperative work-up or to assess down staging in most oncological and surgical populations. The total body is rarely depicted in this setting. Therefore, the CSMA at the level of L3, which accurately estimated total body skeletal muscle mass [4, 31] proved its use. CT is currently the golden standard for body composition measurements [32] with excellent inter- en intra-observer agreement [13]. Some studies advocate that CSMA is superior to psoas muscle measurements alone [33], whereas other studies state otherwise [16]. Besides skeletal muscle mass, visceral and subcutaneous adipose tissue may also be measured on CT [34, 35]. However, these measurements are greatly influenced by some factors, such as the position of the visceral organs [13]. Therefore, we decided not to measure adipose tissue in the current study.

In conclusion, sex, age, and BMI were independently associated with skeletal muscle mass and density measured on CT in over a thousand healthy subjects in a Western-European cohort. Our reference values, which are easily available using the online calculator (supplementary material), may be used to calculate the estimated healthy skeletal muscle mass for individuals in patient populations.

Acknowledgements

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REFERENCES

1. Heymsfield SB, Wang Z, Baumgartner RN, Ross R. Human body composition: advances in models and methods. Annu Rev Nutr 1997; 17: 527-58.
2. Kvist H, Sjostrom L, Tylen U. Adipose tissue volume determinations in women by computed tomography: technical considerations. Int J Obes 1986; 10: 53-67.
3. Prado CM, Birdsell LA, Baracos VE. The emerging role of computerized tomography in assessing cancer cachexia. Curr Opin Support Palliat Care 2009; 3: 269-75.
4. Shen W, Punyanitya M, Wang Z, Gallagher D, St-Onge MP, Albu J, et al. Total body skeletal muscle and adipose tissue volumes: estimation from a single abdominal cross-sectional image. J Appl Physiol (1985) 2004; 97: 2333-8.
5. Prado CM, Lieffers JR, McCargar LJ, Reiman T, Sawyer MB, Martin L, et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. Lancet Oncol 2008; 9: 629-35.
6. Levolger S, van Vugt JL, de Bruin RW, IJzermans JN. Systematic review of sarcopenia in patients operated on for gastrointestinal and hepatopancreatobiliary malignancies. Br J Surg 2015; 102: 1448-58.
7. van Vugt JL, Levolger S, de Bruin RW, van Rosmalen J, Metselaar HJ, IJzermans JN. Systematic review and meta-analysis of the impact of computed tomography assessed skeletal muscle mass on outcome in patients awaiting or undergoing liver transplantation. Am J Transplant 2016.
8. Mourtzakis M, Prado CM, Lieffers JR, Reiman T, McCargar LJ, Baracos VE. A practical and precise approach to quantification of body composition in cancer patients using computed tomography images acquired during routine care. Appl Physiol Nutr Metab 2008; 33: 997-1006.
9. van Vledder MG, Levolger S, Ayez N, Verhoef C, Tran TC, IJzermans JN. Body composition and outcome in patients undergoing resection of colorectal liver metastases. Br J Surg 2012; 99: 550-7.
10. Coelen RJ, Wiggers JK, Nio CY, Besselink MG, Busch OR, Gouma DJ, et al. Preoperative computed tomography assessment of skeletal muscle mass is valuable in predicting outcomes following hepatectomy for perihilar cholangiocarcinoma. HPB (Oxford) 2015; 17: 520-8.
11. Martin L, Birdsell L, Macdonald N, Reiman T, Clandinin MT, McCargar LJ, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol 2013; 31: 1539-47.
12. Masanes F, Rojano ILX, Salva A, Serra-Rexach JA, Artaza I, Formiga F, et al. Cut-off Points for Muscle Mass - Not Grip Strength or Gait Speed - Determine Variations in Sarcopenia Prevalence. J Nutr Health Aging 2017; 21: 825-9.

13. van Vugt JL, Levolger S, de Bruin RW, IJzermans JN. A comparative study of software programs for cross-sectional skeletal muscle area measurements on abdominal computed tomography scans. J Cachexia Sarcopenia Muscle 2015.
14. van Vugt JLA, Coebergh van den Braak RRJ, Schippers HJW, Veen KM, Levolger S, de Bruin RWF, et al. Contrast-enhancement influences skeletal muscle density, but not skeletal muscle mass, measurements on computed tomography. Clin Nutr 2017.
15. Carey EJ, Lai JC, Wang CW, Dasarathy S, Lobach I, Montano-Loza AJ, et al. A multicenter study to define sarcopenia in patients with end-stage liver disease. Liver Transpl 2017; 23: 625-33.
16. Golse N, Bucur PO, Ciacio O, Pittau G, Sa Cunha A, Adam R, et al. A new definition of sarcopenia in patients with cirrhosis undergoing liver transplantation. Liver Transpl 2017; 23: 143-54.
17. Yoshizumi T, Shirabe K, Nakagawara H, Ikegami T, Harimoto N, Toshima T, et al. Skeletal muscle area correlates with body surface area in healthy adults. Hepatol Res 2014; 44: 313-8.
18. Collaboration NCDRF. Trends in adult body-mass index in 200 countries from 1975 to 2014: a pooled analysis of 1698 population-based measurement studies with 19.2 million participants. Lancet 2016; 387: 1377-96.
19. Garatachea N, Lucia A. Genes and the ageing muscle: a review on genetic association studies. Age (Dordr) 2013; 35: 207-33.
20. Welle S, Tawil R, Thornton CA. Sex-related differences in gene expression in human skeletal muscle. PLoS One 2008; 3: e1385.
21. Hamaguchi Y, Kaido T, Okumura S, Kobayashi A, Hammad A, Tamai Y, et al. Proposal for new diagnostic criteria for low skeletal muscle mass based on computed tomography imaging in Asian adults. Nutrition 2016; 32: 1200-5.
22. Baracos VE. Psoas as a sentinel muscle for sarcopenia: a flawed premise. J Cachexia Sarcopenia Muscle 2017; 8: 527-8.
23. Franssen FM, Rutten EP, Groenen MT, Vanfleteren LE, Wouters EF, Spruit MA. New reference values for body composition by bioelectrical impedance analysis in the general population: results from the UK Biobank. J Am Med Dir Assoc 2014; 15: 448 e1-6.
24. Baumgartner RN, Koehler KM, Gallagher D, Romero L, Heymsfield SB, Ross RR, et al. Epidemiology of sarcopenia among the elderly in New Mexico. Am J Epidemiol 1998; 147: 755-63.
25. Thorpe RJ, Simonsick E, Zonderman A, Evans MK. Association between Race, Household Income and Grip Strength in Middle- and Older-Aged Adults. Ethn Dis 2016; 26: 493-500.
26. Guessous I, Luthi JC, Bowling CB, Theler JM, Paccaud F, Gaspoz JM, et al. Prevalence of frailty indicators and association with socioeconomic status in middle-aged and older adults in a swiss region with universal health insurance coverage: a population-based cross-sectional study. J Aging Res 2014; 2014: 198603.

27. Cruz-Jentoft AJ, Baeyens JP, Bauer JM, Boirie Y, Cederholm T, Landi F, et al. Sarcopenia: European consensus on definition and diagnosis: Report of the European Working Group on Sarcopenia in Older People. Age Ageing 2010; 39: 412-23.

28. Popuri K, Cobzas D, Esfandiari N, Baracos V, Jagersand M. Body Composition Assessment in Axial CT Images Using FEM-Based Automatic Segmentation of Skeletal Muscle. IEEE Trans Med Imaging 2016; 35: 512-20.

29. Akhtar-Danesh N, Dehghan M, Merchant AT, Rainey JA. Validity of self-reported height and weight for measuring prevalence of obesity. Open Med 2008; 2: e83-8.

30. Gunnell D, Berney L, Holland P, Maynard M, Blane D, Frankel S, et al. How accurately are height, weight and leg length reported by the elderly, and how closely are they related to measurements recorded in childhood? Int J Epidemiol 2000; 29: 456-64.

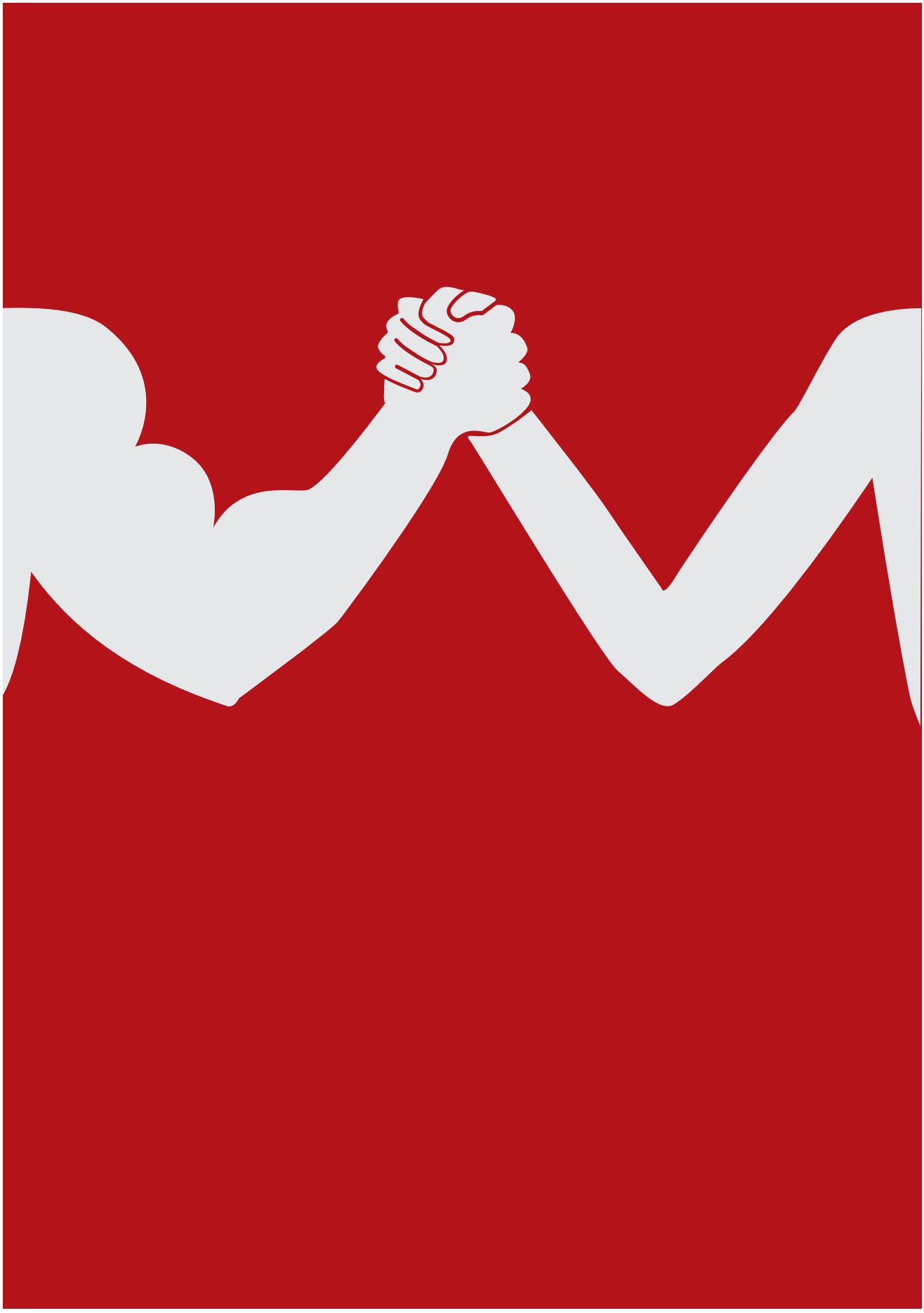
31. Shen W, Punyanitya M, Wang Z, Gallagher D, St-Onge MP, Albu J, et al. Visceral adipose tissue: relations between single-slice areas and total volume. The American journal of clinical nutrition 2004; 80: 271-8.

32. Giusto M, Lattanzi B, Albanese C, Galtieri A, Farcomeni A, Giannelli V, et al. Sarcopenia in liver cirrhosis: the role of computed tomography scan for the assessment of muscle mass compared with dual-energy X-ray absorptiometry and anthropometry. Eur J Gastroenterol Hepatol 2015; 27: 328-34.

33. Rutten IJ, Ubachs J, Kruitwagen RFPM, Beets-Tan RGH, Olde Damink SWM, van Gorp T. Abstracts of the 9th International Conference on Cachexia, Sarcopenia, and Muscle Wasting, Berlin, Germany, 10–11 December 2016 (part 2) (1-45: Psoas muscle measurements are inferior to total skeletal muscle measurements in the assessment of sarcopenia in ovarian cancer). J Cachexia Sarcopenia Muscle 2017; 8: 161-83.

34. Cakir H, Heus C, van der Ploeg TJ, Houdijk AP. Visceral obesity determined by CT scan and outcomes after colorectal surgery; a systematic review and meta-analysis. Int J Colorectal Dis 2015; 30: 875-82.

35. van Grinsven J, van Vugt JL, Gharbharan A, Bollen TL, Besselink MG, van Santvoort HC, et al. The Association of Computed Tomography-Assessed Body Composition with Mortality in Patients with Necrotizing Pancreatitis. Journal of gastrointestinal surgery : official journal of the Society for Surgery of the Alimentary Tract 2017.



PART III

CONSEQUENCES OF LOW SKELETAL MUSCLE MASS IN SURGICAL ONCOLOGY

III

CHAPTER 5

Systematic Review of Sarcopenia in Patients Operated on for Gastrointestinal and Hepatopancreatobiliary Malignancies

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5

ABSTRACT

Background: Preoperative risk assessment in cancer surgery is of importance to improve treatment and outcome. The aim of this study was to assess the impact of CT-assessed sarcopenia on short- and long-term outcomes in patients undergoing surgical resection of gastrointestinal and hepatopancreatobiliary malignancies.

Methods: A systematic search of Embase, PubMed and Web of Science was performed to identify relevant studies published before 30 September 2014. PRISMA guidelines for systematic reviews were followed. Screening for inclusion, checking the validity of included studies and data extraction were carried out independently by two investigators.

Results: After screening 692 records, 13 observational studies with a total of 2884 patients were included in the analysis. There was wide variation in the reported prevalence of sarcopenia (17.0–79 %). Sarcopenia was independently associated with reduced overall survival in seven of ten studies, irrespective of tumor site. Hazard ratios (HRs) of up to 3.19 (hepatic cancer), 1.63 (pancreatic cancer), 1.85 (colorectal cancer) and 2.69 (colorectal liver metastases, CLM) were reported. For esophageal cancer, the HR was 0.31 for increasing muscle mass. In patients with colorectal cancer and CLM, sarcopenia was independently associated with postoperative mortality (colorectal cancer: odds ratio (OR) 43.3), complications (colorectal cancer: OR 0.96 for increasing muscle mass; CLM: OR 2.22) and severe complications (CLM: OR 3.12).

Conclusions: Sarcopenia identified before surgery by single-slice CT is associated with impaired overall survival in gastrointestinal and hepatopancreatobiliary malignancies, and increased postoperative morbidity in patients with colorectal cancer with or without hepatic metastases.

INTRODUCTION

Advanced surgical techniques, developments in perioperative care and the introduction of enhanced recovery programs have improved surgical outcomes [1–5]. Nevertheless, risk assessment before major abdominal surgery remains of paramount importance to further improve outcomes after cancer surgery. Known factors that are predictive of short-term outcome include albumin levels, American Society of Anesthesiologists (ASA) classification and emergency surgery, whereas advanced age and disseminated disease determine long-term outcome [6–8]. Outcomes of patients with similar age, tumor stage and ASA classification may be very different in clinical practice. Therefore, the risk factors commonly used to predict outcome after cancer surgery may reflect the patient's general health status and physiological reserves insufficiently. An important risk factor for worse outcome is frailty, which is poorly reflected by the traditional determinants of outcome [9–13]. Frailty is defined as a biological syndrome characterized by decreased reserve and resilience to stress factors across multiple physiological systems, and has been shown to be associated with adverse health outcomes [14,15]. A hallmark sign of frailty is sarcopenia, the involuntary loss of skeletal muscle mass [16–18]. The prevalence of sarcopenia in healthy individuals increases with advanced age, ranging from 9 % at 45 years and up to 64 % in individuals aged over 85 years [19].

Sarcopenia is characterized by a loss of skeletal muscle mass, skeletal muscle strength and physical performance [20]. It has been shown to impair physical performance and survival in geriatric, non-cancer populations [21,22], and to impair survival in a variety of clinical conditions, such as cancer [23]. Up to 80 % of patients with advanced cancer are affected by cancer-induced cachexia, a clinical condition that also results in skeletal muscle wasting with or without loss of body fat [24–26]. Cachectic patients are more prone to a reduced effect of therapy and increased chemotherapy toxicity [27–29]. It has been estimated that as many as 30 % of cancer-related deaths result from cachexia [30–33]. One study [23] showed that sarcopenia was associated with decreased survival in obese patients with cancer by using CT to assess reduced skeletal muscle mass [34] (figure 1).

A systematic review was undertaken to investigate the influence of low skeletal muscle mass or skeletal muscle density assessed by CT on short- and long-term outcomes in patients undergoing surgery for gastrointestinal and hepatopancreatobiliary malignancies.

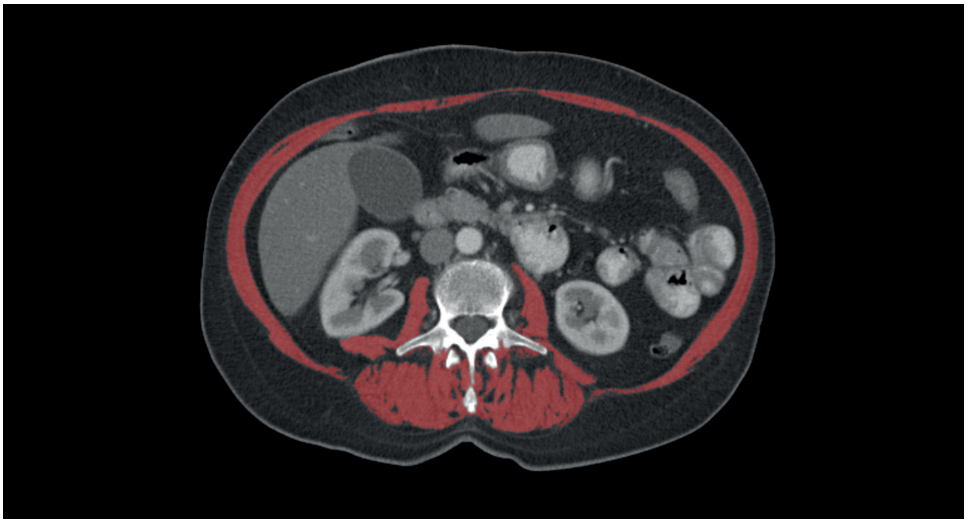

Figure 1. Transverse CT image at the level of L3 showing a cross-sectional area of skeletal muscle mass highlighted in red, including the psoas, paraspinal, transverse abdominal, external oblique, internal oblique and rectus abdominis muscles.

METHODS

Eligibility criteria were established *a priori*. A systematic search was performed to identify all original articles on patients undergoing surgical resection of malignancies of the gastrointestinal tract or hepatopancreatobiliary system, in which preoperative abdominal CT was used to assess skeletal muscle mass. The Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines were followed [35].

Included in the analysis were studies that reported on the prevalence of sarcopenia, and at least one of the following outcomes: postoperative mortality, postoperative complications, length of intensive care (ICU) stay, length of hospital stay, disease-free survival and overall survival.

The search was limited to papers in English with a publication date from January 2000 to September 2014. Three search strings with corresponding search terms were constructed (table S1, supporting information). The same search strings were used to develop queries in the EMBASE, PubMed and Web of Science databases.

The EMBASE database search was performed using the following query: ('sarcopenia':de,ab,ti OR 'analytic morphomics':de,ab,ti OR 'body composition':de,ab,ti OR 'muscle depletion':de,ab,ti OR 'muscle mass':de,ab,ti OR 'psoas area':de,ab,ti OR 'myopenia':de,ab,ti OR 'core muscle':de,ab,ti OR 'lean body mass':de,ab,ti OR 'muscular atrophy':de,ab,ti) AND ('cancer':de,ab,ti OR 'neoplasms':de,ab,ti OR 'malignancy':de,ab,ti) AND ('surgery':de,ab,ti OR 'resection':de,ab,ti OR 'esophagectomy':de,ab,ti OR 'gastrectomy':de,ab,ti OR 'hepatectomy':de,ab,ti OR 'colectomy':de,ab,ti OR 'pancreatectomy':de,ab,ti or 'cholecystectomy':de,ab,ti). Similar queries were constructed for PubMed and Web of Science.

Duplicate records were removed and abstracts screened independently by two investigators to determine which records were eligible for further analysis. Abstracts were included for initial analysis if sarcopenia in patients undergoing surgical treatment with gastrointestinal or hepatopancreatobiliary malignancies was described. Abstracts that described sarcopenia determined by means other than abdominal CT or patients undergoing non-surgical treatment were excluded from further analysis. Records without abstracts, case reports, review articles, opinion articles and experimental studies were excluded.

Eligibility of studies and assessment of methodological quality

Full-text articles of the remaining records were subsequently retrieved and screened independently by two investigators. All original articles that met the inclusion criteria were included. Additional relevant references were sought in the included full-text articles. Two investigators independently assessed the methodological quality of the included studies using the Newcastle–Ottawa quality assessment scale for cohort studies [36] for each *a priori* defined outcome measure.

Data extraction

Data regarding study design and results were extracted independently by two investigators for each eligible study. Extracted data included age, sex distribution, patient selection, prevalence of sarcopenia, postoperative mortality, postoperative complications, length of ICU stay, length of hospital stay, disease-free survival and overall survival. If univariable and multivariable analyses had been performed to adjust for known risk factors, the latter was used for interpretation of the results.

Statistical analysis

Outcomes are reported as originally shown. The prevalence of sarcopenia described in this review applies to the total population of each study. Therefore, rates could not be provided for subgroups (such as by cancer stage) separately. No meta-analysis was performed because there was great heterogeneity between studies.

RESULTS

The literature search was performed on 30 September 2014 and identified an initial 692 records, of which 27 were found to be potentially relevant (figure 2). From these 27 records, seven full-text articles were excluded as sarcopenia was assessed by means other than abdominal CT, four articles did not report relevant outcome data, and three articles reported on a population that received non-surgical treatment for the studied tumors. The remaining 13 studies matched the inclusion criteria [37–49]. Cross-referencing yielded no additional results. The included studies provided data on patients with esophageal, gastric, pancreatic, primary liver and colorectal cancer, and resectable hepatic colorectal metastases (table 1). No studies reported on patients with bile duct or gallbladder cancer.

Prevalence of sarcopenia in different malignancies

The prevalence of sarcopenia as assessed by CT-based skeletal muscle mass measurement in patients undergoing surgery for gastrointestinal and hepatopancreatobiliary malignancies was reported in nine studies [37–45,47]. None of the studies [39,41,42,44,45,49] that compared characteristics in patients with and without sarcopenia reported on significant differences regarding cancer stage, differentiation grade or biomarkers. Despite comparable age and sex distribution between studies, there was a wide variation in the prevalence of sarcopenia, ranging from 17.0% in a cohort of patients with hepatic colorectal metastases [45] to 79 % in a cohort with esophageal and gastric cancer [37]. In agreement, cohorts of patients with esophageal and gastric cancer reported a widespread prevalence of sarcopenia before surgery, ranging from 43 to 79% [37,38]. Less variation in the prevalence of sarcopenia was observed among patients undergoing surgical resection of hepatocellular carcinoma (40.3–54.1%) [39–41], colorectal cancer (38.9–47.7%) [42,43] and hepatic colorectal metastases (17.0–19.4%) [44,45]. One study [47] reported a prevalence of sarcopenia of 25.0% in patients with pancreatic cancer. Two studies [37,38] reported an increase in the prevalence of sarcopenia among patients with esophageal and gastric cancer following neoadjuvant chemotherapy. The impact of neoadjuvant therapy on the prevalence of sarcopenia was not assessed in the colorectal cancer studies included in the present analysis. A possible impact of age or sex on the prevalence of sarcopenia could not be discerned. Detailed information regarding the prevalence of sarcopenia is shown in table 2.

Table 1. Studies of the effects of sarcopenia in patients operated for gastrointestinal and hepatopancreatobiliary malignancies.

							Quality points by outcome*		
Reference	**Malignancy, patient selection**	**Disease stage (%)**	***n* (Men)**	**Age (years)**	**BMI (kg/m²)**	**Muscle(s) measured (level), cut-offs**	**Short-term morbidity or mortality**	**DFS**	**OS**
Awad *et al.*[37] (2012)	Esophageal and gastric cancer, WHO performance status 0–2	Locally advanced	47 (34)	63†	Before NACRT: 24.6† Before resection: 23.8†	CSAMM/m² (L3) F 38.5 cm² M 52.4 cm²	5	n.r.	4
Sheetz *et al.*[46] (2013)	Esophageal cancer, all consecutive patients	IS: 9.6 I: 24.3 II: 33.5 III: 27.4 IV: 5.2	230 (202)	62†	Overall: 28.6†	TPA, PMD (L4) –	7	6	7
Yip *et al.*[38] (2014)	Esophageal cancer	IS: 6 I: 3 II: 51 III: 40	35 (30)	63†	Before NACRT: 26.7† Before resection: 25.8†	CSAMM/m² (L3) F 38.5 cm² M 52.4 cm²	5	5	5
Harimoto *et al.*[41] (2013)	Hepatocellular cancer,all consecutive patients	I: 15.6 II: 51.1 III: 26.3 IV: 7.0	186 (145)	–	Sarcopenia: 20.5† No sarcopenia: 24.0†	CSAMM/m² (L3) F 41.1 cm² M 43.75 cm²	7	6	6
Itoh *et al.*[40] (2014)	Hepatocellular cancer, all patients without simultaneous procedures	n.a.	190 (146)	–	<18.5: 7.9% ≥18.5 to < 25: 68.4% ≥ 25 to < 30: 21.6% ≥ 30: 2.1%	CSAMM/m² (L3) F 41.1 cm² M 43.75 cm²	n.r.	6	6
Voron *et al.*[39] (2014)	Hepatocellular cancer, all consecutive patients	n.a.	109 (92)	62†	Overall: 24.6† Sarcopenia: 25.6† Noarcopenia: 26.9†	CSAMM/m² (L3) F 38.9 cm² M 52.4 cm²	7	6	6
Peng *et al.*[47] (2012)	Pancreatic cancer, all consecutive patients	IS: 0.2 I: 5.9 II: 16.9 III: 71.5 IV: 4.0 n.a.: 1.6	557 (296)	66†	≥ 30: 20.1%	TPA (L3) F 362 mm²/m² M 492 mm²/m²	6	n.r.	5

Reference	Malignancy, patient selection	Disease stage (%)	*n* (Men)	Age (years)	BMI (kg/m²)	Muscle(s) measured (level), cut-offs	Quality points by outcome*		
							Short-term morbidity or mortality	DFS	OS
Jung *et al.*[49] (2014)	Colorectal cancer	All stage III receiving adjuvant chemotherapy	229 (134)	61‡	Sarcopenia: 22.2† (< 30: 87.8%) No sarcopenia: 23.6† (< 30: 71.1%)	TPA/m² (L4) –	n.r.	7	7
Lieffers *et al.*[42] (2012)	Colorectal cancer	II: 31.6 III: 35.5 IV: 32.9	234 (135)	63†	Overall: 28.5† Sarcopenia: 26.1† No sarcopenia: 30.0†	CSAMM/m² (L3) F 38.5 cm² M 52.4 cm²	6	n.r.	n.r.
Reisinger *et al.*[43] (2014)	Colorectal cancer, all consecutive patients	I–II: 46.7 III–IV: 53.3	310 (155)	69†	> 25: 58.7%	CSAMM/m² (L3) F 38.5 cm² M 52.4 cm²	7	n.r.	n.r.
Sabel *et al.*[48] (2013)	Colorectal cancer, all consecutive patients	I: 24 II: 33 II: 30 IV: 11 n.a.: 2	302 (157)	68†	Overall: 28.7†	PMD (L4) –	7	7	7
Peng *et al.*[45] (2011)	Colorectal liver metastases, all consecutive patients	All stage IV	259 (155)	58‡	≥ 30: 26.0%	TPA/m² (L3) 500 mm²	6	5	5
van Vledder *et al.*[44] (2012)	Colorectal liver metastases, all consecutive patients	All stage IV	196 (120)	65‡	Sarcopenia: 23.7† No sarcopenia: 26.7†	CSAMM/m² (L3) F 41.1 cm² M 43.75 cm²	n.r.	7	7

*Score from a maximum of 9 using the Newcastle–Ottawa quality assessment scale for cohort studies. †Mean;‡median. BMI, body mass index; DFS, disease-free survival; OS, overall survival; WHO, World Health Organization; NACRT, neoadjuvant chemotherapy; CSAMM, cross-sectional area of muscle mass; m², squared body height; L3/4, at the level of the third/fourth lumbar vertebra; IS, *in situ;* TPA, total psoas area; PMD, psoas mean density; n.a., not available; n.r., not recorded.

Table 2. Studies reporting the prevalence of sarcopenia in gastrointestinal malignancies.

Reference	Malignancy	Prevalence (%)
Awad *et al.*[37]	Esophageal and gastric cancer	Before NACRT: 57 | Before resection: 79
Yip *et al.*[38]	Esophageal cancer	Before NACRT: 26 | Before resection: 43
Voron *et al.*[39]	Hepatocellular carcinoma	54.1
Itoh *et al.*[40]	Hepatocellular carcinoma	40.5
Harimoto *et al.*[41]	Hepatocellular carcinoma	40.3
Peng *et al.*[47]	Pancreatic cancer	25.0
Lieffers *et al.*[42]	Colorectal cancer	38.9
Reisinger *et al.*[43]	Colorectal cancer	47.7
van Vledder *et al.*[44]	Colorectal liver metastases	19.4
Peng *et al.*[45]	Colorectal liver metastases	17.0

NACRT, neoadjuvant chemotherapy.

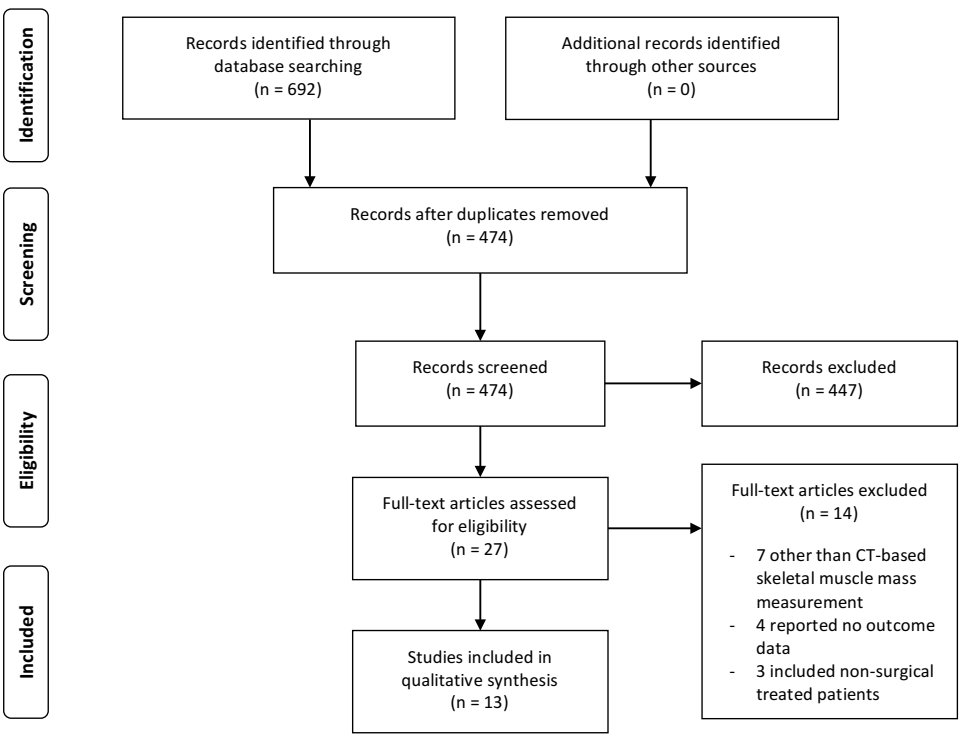

Figure 2. PRISMA flow chart showing selection of articles for review.

Short-term postoperative morbidity and mortality

Data regarding complication rate, length of ICU stay, length of hospital stay, postoperative morbidity and postoperative mortality were reported in ten [37–39,41–43,45–48] of the 13 studies included in the analysis (table 3).

An increased postoperative morbidity rate was found in patients with sarcopenia in all studies where this was reported among patients undergoing surgical resection of colorectal cancer [42,43,48] and hepatic colorectal metastases [45]. One study [48] reported that an increase in psoas density protected against overall (odds ratio (OR) 0.96, 95% c.i. 0.94 to 0.99; p=0.004) and infectious (OR 0.95, 0.93 to 0.98; p=0.001) complications in a cohort of 302 patients [48]. Another investigation [42] observed an increase in infectious complications in patients with versus those without sarcopenia (23.1 versus 12.6%; p=0.036) in a cohort of 234 patients. Subgroup analysis revealed that the risk was especially pronounced in elderly patients (65 years or older) with sarcopenia (29.6 versus 8.8%; p=0.005). This difference remained significant in multivariable analysis (adjusted OR 4.6, 1.5 to 13.9; p=0.007). The overall complication rate was not described. An increased risk of major postoperative complications (Clavien–Dindo grade IIIa or higher) among patients with sarcopenia compared with those without was reported among patients undergoing hepatic resection for colorectal metastases (22 versus 8% respectively; OR 3.12; p=0.020) [45]. However, the study did not specify the type of complications. Another investigation [43] showed a strong association between sarcopenia and 30-day mortality combined with in-hospital mortality after elective colorectal cancer surgery (8.8 versus 0.6% in patients with and without sarcopenia respectively; OR 43.3, 2.74 to 685.2, p=0.007).

No association between sarcopenia and postoperative morbidity and mortality was found in patients undergoing resection for esophageal or hepatocellular cancer [37,39,41,46]. Specifically, in a cohort of 557 patients undergoing pancreatic cancer resection [47], there was no difference in the rate of any postoperative complication (44.6 versus 51.8% in men with and without sarcopenia respectively, p=0.28, 41.5 versus 43.4% respectively among women, p=0.80), major postoperative complications (20.6 versus 24.8% for men, p=0.49; 12.1 versus 20.5% for women, p=0.15) or 30-day postoperative mortality (1.4 versus 0.5% for men, p=0.44; 0 versus 0.5% for women, p=1.00). However, the 90-day mortality rate differed between men with and without sarcopenia (9.5 versus 2.7% respectively; p=0.02).

Table 3. Studies reporting the impact of sarcopenia on short-term outcome in patients operated for gastrointestinal malignancies.

		Complications					Length of stay (days)	
Reference	**Malignancy**	**All**	**Clavien–Dindo classification ≥ IIIa**	**Postop./in-hospital mortality**	**Anastomotic leakage**	**Infectious**	**ICU**	**Hospital**
Awad *et al.*[37]*	Esophageal and gastric cancer			p=0.060				p=0.51
Sheetz *et al.*[46]	Esophageal cancer	LPA: 1993 *versus* 1877 mm^2, without *versus* with complications (p=0.12)			LPA: 1922 *versus* 1953 mm^2, no leakage *versus* leakage (p=0.40)			
Yip *et al.*[38]	Esophageal cancer	n.s.						n.s.
Harimoto *et al.*[41]	Hepatocellular carcinoma	32.0 *versus* 50.5% (p=0.61)						
Voron *et al.*[39]	Hepatocellular carcinoma	39.0 *versus* 36.0% (p=0.749)	20.3 *versus* 16.0% (p=0.560)	6.8 *versus* 2.0% (p=0.372)				
Peng *et al.*[47]	Pancreatic cancer	OR 0.88 (0.60,1.29) (p=0.51)	OR 0.72 (0.43, 1.21) (p=0.21)	HR 2.31 (0.78, 6.77) (p=0.13)			0.4 *versus* 0.4 (p=0.92)	12 *versus* 12 (p=0.98)
Lieffers *et al.*[42]	Colorectal cancer					23.1 *versus* 12.6% OR 4.6 (p=0.007)†		15.9 *versus* 12.3 (p=0.038)
Reisinger *et al.*[43]	Colorectal cancer			OR 43.3 (2.74, 685.2) (p=0.007)†	OR 0.57 (0.28, 1.19) (p=0.13)			
Sabel *et al.*[48]	Colorectal cancer	OR 0.96 (0.94, 0.99) for every unit of increased psoas density (p=0.004)†				OR 0.95 (0.93, 0.98) for every unit of increased psoas density (p=0.001)†		
Peng *et al.*[45]	Colorectal liver metastases	OR 2.22 (p=0.02)	22 *versus* 8% OR 3.12 (1.14, 8.49) (p=0.02)†				Prolonged stay (> 2 days): 15 *versus* 4% (p=0.004)	6.6 *versus* 5.4 (p=0.03)

Data are shown for groups with *versus* without sarcopenia unless indicated otherwise. Odds ratios (ORs) and hazard ratios (HRs) are shown with 95 % c.i. *Using fat-free mass assessed by CT before resection. †Multivariable analysis. ICU, intensive care unit; LPA, lean psoas area; n.s., not significant.

Two studies [43,46] that reported on anastomotic leakage following surgical resection of colorectal and esophageal cancer did not demonstrate an association with sarcopenia.

Two studies adjusted for body mass index (BMI) in the multivariable analyses. One [43] reported that sarcopenia was a risk factor for 30-day mortality, whereas BMI was not. Similarly, in another investigation [45] sarcopenia, but not BMI, was a risk factor for postoperative complications.

Length of intensive care unit and hospital stay

Peng and colleagues reported a prolonged ICU admission (more than 2 days) for patients with sarcopenia undergoing resection with curative intent for hepatic colorectal metastases compared with those without sarcopenia (15 versus 4% respectively; p=0.004) [45], but did not demonstrate a difference in the mean length of ICU stay in patients undergoing surgical resection of pancreatic cancer (mean (s.d.) 0.5(2.0) versus 0.5(1.7) days respectively for men, p=1.00; 0.2(0.6) versus 0.2(0.6) days among women, p=0.74) [47].

In two [42,45] of five studies [37,38,42,45,47] reporting length of hospital stay, patients with sarcopenia had a delayed discharge from hospital. Hospital stay was slightly prolonged in patients with sarcopenia undergoing resection with curative intent for hepatic colorectal metastases (6.6 versus 5.4 days; p=0.03) [45]. The impact of sarcopenia on length of hospital stay may be greater in conjunction with other patient characteristics. For instance, hospital stay was significantly longer in patients with sarcopenia than in those without for all patients undergoing surgery for colorectal cancer (15.9 versus 12.3 days; p=0.038). The corresponding rates for patients aged 65 years or older were 20.2 versus 13.1 days (p=0.008). In addition, sarcopenia was an independent factor for the need for rehabilitation in patients aged 65 years and older (OR 3.1, 95% c.i. 1.4 to 9.4; p<0.040) [42]. The two studies [42,45] that reported an increased length of hospital stay in patients with sarcopenia also observed an increased number of postoperative complications. Length of hospital stay did not significantly differ between patients with and without sarcopenia in studies of pancreatic cancer [47] and esophageal and gastric cancer [37,38],

Disease-free survival

Nine studies [38–41,44–46,48,49] described the association between sarcopenia and disease-free survival. Data regarding disease-free survival rates and times in the individual studies are shown in table 4 and figure 3.

Table 4. Studies reporting the impact of sarcopenia on long-term outcomes in gastrointestinal malignancies.

Reference	Malignancy	Disease-free survival	Overall survival
Awad *et al.*[37]	Esophageal and gastric cancer	n.r.	12-month mortality equal in patients with low FFM and those with normal FFM after NACRT (p=0.57)
Sheetz *et al.*[46]	Esophageal cancer	NACRT: HR 0.83 (0.52,1.33) for increasing LPA (p=0.433)* No NACRT: HR 0.33 (0.14, 0.80) for increasing LPA (p=0.014)*	NACRT: HR 0.77 (0.46, 1.28) for increasing LPA (p=0.311)* No NACRT: HR 0.31 (0.12, 0.82) for increasing LPA (p=0.018)*
Yip *et al.*[38]	Esophageal cancer	n.s.	After chemotherapy: Median 25.6 months *versus* median not reached (p=0.063)
Itoh *et al.*[40]	Hepatocellular carcinoma	HR 1.30 (0.85, 2.00) (p=0.215)*	HR 1.96 (1.06, 2.83) (p=0.031)*
Harimoto *et al.*[41]	Hepatocellular carcinoma	5-year: 13.0 *versus* 33.2 (p=0.013) HR 0.97 (0.95, 1.00) for increasing muscle mass (p=0.016)*	5-year: 71 *versus* 83.7% (p=0.001) HR 0.90 (0.84, 0.96) for increasing muscle mass (p=0.002)*
Voron *et al.*[39]	Hepatocellular carcinoma	HR 3.03 (1.67, 5.49) (p<0.001)*	HR 3.19 (1.28, 7.96) (p=0.013)*
Peng *et al.*[47]	Pancreatic cancer	n.a.	M, 3-year: 20.3 *versus* 39.2% (p<0.050) F, 3-year: 26.1% *versus* 40.8 (p<0.050) 3-year: HR 1.63 (1.28, 2.07) (p<0.001)*
Jung *et al.*[49]	Colorectal cancer	p=0.946*	HR 1.85 (1.10, 3.13) (p=0.022)*
Sabel *et al.*[48]	Colorectal cancer	HR 0.97 (0.95, 1.00) (p=0.03) for increasing PD n.s.*	HR 0.97 (0.95, 1.00) for increasing PD (p=0.04) n.s.*
Peng *et al.*[45]	Colorectal liver metastases	HR 1.07 (p=0.78)	HR 1.05 (p=0.80)
van Vledder *et al.*[44]	Colorectal liver metastases	HR 1.96 (1.29, 2.97) (p=0.002)*	HR 2.69 (1.67, 4.32) (p<0.001)*

Data are shown for groups with *versus* without sarcopenia unless indicated otherwise. Hazard ratios (HRs) are shown with 95 % c.i. *Multivariable analysis. NACRT, neoadjuvant chemoradiotherapy; LPA, lean psoas area; n.s., not significant; n.a., not available; PD, psoas density.

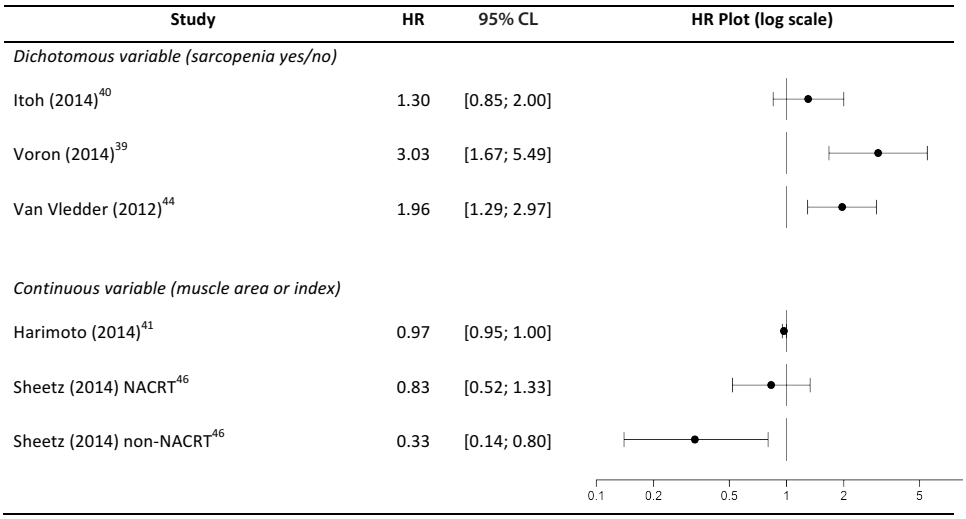

Figure 3. Forest plots showing studies that reported disease-free survival. Only studies reporting hazard ratios with 95 % c.i. are shown. NACRT, neoadjuvant chemoradiotherapy.

In patients with esophageal cancer, sarcopenia was associated with impaired disease-free survival in those who underwent surgical resection without receiving neoadjuvant chemoradiotherapy independently of age, sex and tumor stage (hazard ratio (HR) 0.33, 95% c.i. 0.14 to 0.80; p=0.014) [46]. However, no association between sarcopenia and disease-free survival was observed in patients who underwent surgical resection following neoadjuvant chemoradiotherapy [38,46].

Patients with hepatocellular cancer who had sarcopenia had an increased risk of disease recurrence in two [39,41] of three [39–41] studies. One study [39] reported a median disease-free survival of 10.1 months in patients with sarcopenia and 34.2 months in those without sarcopenia (p<0.001), and an independent association between sarcopenia and disease-free survival (HR 3.03, 95% c.i. 1.67 to 5.49; p<0.001). Another study [41] reported 5-year disease-free survival rates in patients with and without sarcopenia of 13.0 and 33.2% respectively (p=0.013). In multivariable analysis, a high skeletal muscle mass was independently associated with a lower risk of disease recurrence (HR 0.97, 0.95 to 1.00; p=0.016). Yet another study [40] reported reduced disease-free survival in patients undergoing hepatocellular cancer resection in univariable analysis (HR 1.62, 1.11 to 2.36; p=0.012), but this association did not remain significant in the multivariable analysis (HR 1.30, 0.85 to 2.00; p=0.215).

In patients with primary colorectal cancer, sarcopenia impaired disease-free survival in one [48] of the two studies [48,49] reporting on disease recurrence. One study [48] described a protective effect of high psoas muscle density (HR 0.97, 0.95 to1.00; p=0.03). However, there was no significant difference in disease-free survival between patients with normal and low skeletal muscle mass in another study [49]. No median survival times, or 1-, 3- or 5-year survival rates were reported.

In patients with hepatic colorectal metastases, one study [44] reported a median disease-free survival time of 8.7 months in patients with sarcopenia compared with 15.1 months in patients without sarcopenia (HR 1.96, 1.29 to 2.97; p=0.002). However, another investigation [45] found no association between sarcopenia and disease-free survival in patients with hepatic colorectal metastases; the 5-year recurrence-free survival rate was 23 and 27% in patients with and without sarcopenia respectively (p=0.78).

Five studies [39–41,44,49] made an adjustment for BMI in the analysis of the prognostic value of sarcopenia for disease-free survival. Whereas sarcopenia was associated with disease-free survival in four of nine studies, no association between BMI and disease-free survival was reported in patients with hepatocellular cancer, colorectal cancer or hepatic colorectal metastases.

Overall survival

Most authors reported a significant decrease in overall survival in patients with sarcopenia compared with those without sarcopenia. This effect was observed irrespective of cancer site or tumor origin [39–41,44,46-49]. Data regarding survival rates and median survival times in the individual studies are shown in table 4 and figure 4.

Among patients with esophageal cancer, a trend towards decreased survival among those with sarcopenia was reported in one study (median overall survival 25.6 months versus median not reached for patients without sarcopenia; p=0.063) [38]. In another study [46], overall survival was impaired in patients who had esophageal cancer with sarcopenia and did not receive neoadjuvant chemotherapy (HR 0.31, 95% c.i. 0.12 to 0.82; p=0.018), whereas no significant association was found among patients who did receive neoadjuvant chemotherapy (HR 0.77, 0.46 to 1.28; p=0.311).

A study [39] among patients with hepatocellular carcinoma reported a median survival time of 52.3 and 70.3 months in patients with and without sarcopenia respectively (p=0.015), with a remarkably impaired 1-year survival rate (69.8 versus 95.5%; p=0.015). Another investigation [41] described a less severe impact of sarcopenia on survival in patients with hepatocellular carcinoma, with a reduction in 5-year survival rate from

83.7 to 71% (p=0.001). In a study[47] of patients who had surgery for pancreatic cancer, the 3-year survival rate was lower in patients with sarcopenia than in those without (20.3 versus 39.2% in men, p=0.003; 26.1 versus 40.8% in women, p=0.03). In the multivariable analysis, sarcopenia remained independently associated with an increased risk of death at 3 years (HR 1.63, 1.28 to 2.07; p<0.001).

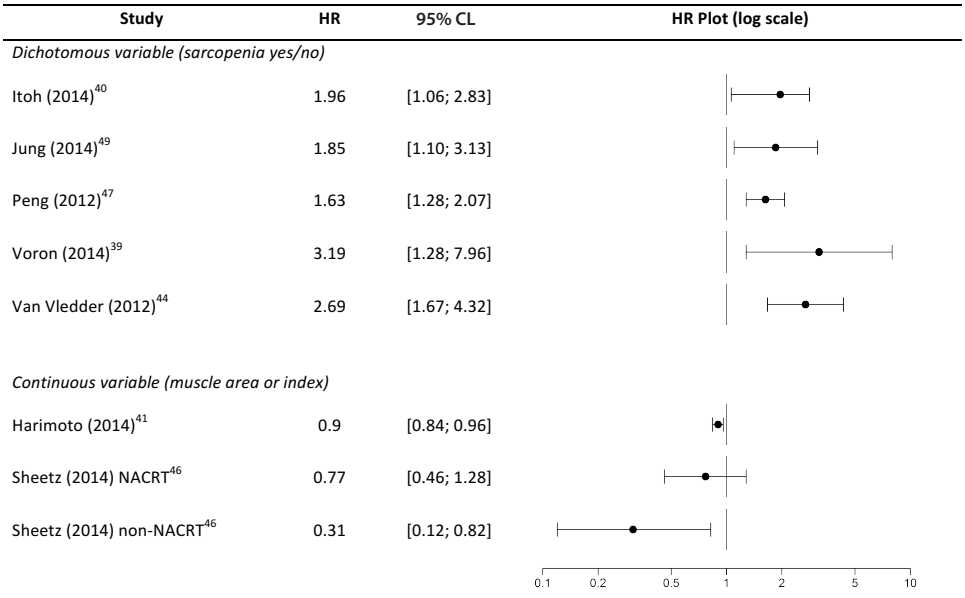

Figure 4. Forest plots showing studies that reported the overall survival. Only studies reporting hazard ratios with 95 % c.i. are shown. NACRT, neoadjuvant chemoradiotherapy.

Median overall survival times, or 1-, 3- or 5-year survival rates were not reported for patients with colorectal cancer in any of the included studies. In patients with hepatic colorectal metastases, one study [44] reported a median survival time of 23.8 versus 59.8 months for patients with and without sarcopenia (HR 2.69, 1.67 to 4.32; p<0.001). Two studies [38,45] found a decreased overall survival in patients with sarcopenia in univariable but not in multivariable analyses.

Five studies undertook multivariable analysis in which the predictive effect of sarcopenia on overall survival was adjusted for BMI. Sarcopenia was independently associated with overall survival in seven of ten studies, whereas no association between BMI and overall survival was reported in patients with hepatocellular cancer and hepatic colorectal

metastases [39–41,44]. However, one study [49] found that a BMI of 25 kg/m^2 or higher was a risk factor for impaired overall survival independent of sarcopenia in patients with stage III colorectal cancer who received adjuvant chemotherapy.

DISCUSSION

Several conclusions can be drawn from this systematic review of the impact of CT-assessed sarcopenia on short- and long-term outcomes in resectable gastrointestinal and hepatopancreatobiliary malignancies. Sarcopenia decreased overall survival, and increased recurrence rates following surgical resection in patients with hepatic colorectal metastases and hepatocellular cancer. Patients with sarcopenia undergoing surgery for colorectal cancer and hepatic colorectal metastases also had a prolonged length of stay after surgery. Because of the heterogeneity of the included studies, the possible influence of age and sex on the prevalence of sarcopenia could not be assessed.

A previous review [52] described the relationship between CT-assessed core muscle size and mortality, postoperative morbidity and length of stay after major abdominal surgery. This systematic review included eight retrospective cohort studies, of which five investigated outcomes in oncological populations. As in the present investigation, sarcopenia was associated with increased morbidity, length of hospital stay and mortality. The relationship between sarcopenia and recurrence was not described.

Preoperative risk stratification is of utmost importance in patient selection for surgery, as it may help physicians to identify patients with a high risk of worse outcome after surgery. A tool suitable for risk evaluation should be inexpensive, easily obtainable and reliable. Bioelectrical impedance analysis, dual-energy X-ray absorptiometry and skinfold measurement are often not performed routinely during the oncological evaluation, whereas the majority of patients undergo abdominal CT as part of preoperative investigations. Cross-sectional muscle area can be measured rapidly by single-slice analysis of abdominal CT images, and is linearly related to total body skeletal muscle mass [53]; this measurement has a low level of interobserver variability [43,53]. CT-based skeletal muscle mass measurement in patients with cancer may identify those at an early stage of frailty, which would otherwise have been undetected clinically [54].

It is still unknown whether treatment of sarcopenia may improve outcomes. Understanding of muscle wasting in cancer has greatly increased over the past decade [55,56] and has led to new treatment options, such as myostatin inhibitors [57,58]. A phase II clinical trial on the efficacy of myostatin inhibitors in patients with advanced or metastatic pancreatic cancer receiving chemotherapy is ongoing, with overall survival as the primary endpoint. There are, however, several other ongoing clinical trials investigating stabilization or reversal of muscle loss in patients with cancer [59].

The present study has some limitations. The included studies were heterogeneous in design, and were predominantly retrospective, observational studies which precluded meta-analysis of the results. Consequently, no causative relationship between sarcopenia and outcome could be demonstrated. Furthermore, the present investigation is likely to have been influenced by submission or publication bias. As there is no standard definition of CT-based assessment of muscle mass, different methods were used, which also hampered evaluation of the results. Investigations that measured total cross-sectional area of muscle mass used distinct sex-specific cut-off values [23,44]. These cut-off values were obtained using the same method of stratification in two different patient populations, yielding two distinct sets of cut-off values. As such, these values may not be interchangeable and applicable to all populations. Another recent study [60] developed a third set of cut-off values, which were both sex- and BMI-specific; these included muscle attenuation, based on Hounsfield units, as a marker for fat infiltration of muscle. These cut-off values remain to be validated.

Sarcopenia impairs overall survival, and may increase postoperative morbidity. Muscle mass assessment by CT may assist in preoperative decision-making, particularly for patients who are thought to be unfit for surgery or who have a high risk of complications.

REFERENCES

1. Kehlet H and Wilmore DW. Multimodal strategies to improve surgical outcome. Am J Surg. 2002;183:630-41.
2. Varadhan KK, Neal KR, Dejong CH, et al. The enhanced recovery after surgery (ERAS) pathway for patients undergoing major elective open colorectal surgery: a meta-analysis of randomized controlled trials. Clin Nutr. 2010;29:434-40.
3. Spanjersberg WR, Reurings J, Keus F, et al. Fast track surgery versus conventional recovery strategies for colorectal surgery. Cochrane Database Syst Rev. 2011;CD007635.
4. Kehlet H. Fast-track colorectal surgery. Lancet. 2008;371:791-3.
5. Zhuang CL, Ye XZ, Zhang XD, et al. Enhanced recovery after surgery programs versus traditional care for colorectal surgery: a meta-analysis of randomized controlled trials. Dis Colon Rectum. 2013;56:667-78.
6. Khuri SF, Henderson WG, DePalma RG, et al. Determinants of long-term survival after major surgery and the adverse effect of postoperative complications. Ann Surg. 2005;242:326-41; discussion 341-3.
7. Kirchhoff P, Clavien PA, and Hahnloser D. Complications in colorectal surgery: risk factors and preventive strategies. Patient Saf Surg. 2010;4:5.
8. Pedersen T. Complications and death following anaesthesia. A prospective study with special reference to the influence of patient-, anaesthesia-, and surgery-related risk factors. Dan Med Bull. 1994;41:319-31.
9. Tan KY, Kawamura YJ, Tokomitsu A, et al. Assessment for frailty is useful for predicting morbidity in elderly patients undergoing colorectal cancer resection whose comorbidities are already optimized. Am J Surg. 2012;204:139-43.
10. Saxton A and Velanovich V. Preoperative frailty and quality of life as predictors of postoperative complications. Ann Surg. 2011;253:1223-9.
11. DePinho RA. The age of cancer. Nature. 2000;408:248-54.
12. Papamichael D, Audisio RA, Glimelius B, et al. Treatment of colorectal cancer in older patients: International Society of Geriatric Oncology (SIOG) consensus recommendations 2013. Ann Oncol. 2014.
13. Hamaker ME, Prins MC, Schiphorst AH, et al. Long-term changes in physical capacity after colorectal cancer treatment. J Geriatr Oncol. 2014.
14. Fried LP, Tangen CM, Walston J, et al. Frailty in older adults: evidence for a phenotype. J Gerontol A Biol Sci Med Sci. 2001;56:M146-56.
15. Milte R and Crotty M. Musculoskeletal health, frailty and functional decline. Best Pract Res Clin Rheumatol. 2014;28:395-410.
16. Cooper C, Dere W, Evans W, et al. Frailty and sarcopenia: definitions and outcome parameters. Osteoporos Int. 2012;23:1839-48.

17. Roubenoff R. Sarcopenia: a major modifiable cause of frailty in the elderly. J Nutr Health Aging. 2000;4:140-2.

18. Marzetti E and Leeuwenburgh C. Skeletal muscle apoptosis, sarcopenia and frailty at old age. Exp Gerontol. 2006;41:1234-8.

19. Cherin P, Voronska E, Fraoucene N, et al. Prevalence of sarcopenia among healthy ambulatory subjects: the sarcopenia begins from 45 years. Aging Clin Exp Res. 2014;26:137-46.

20. Cruz-Jentoft AJ, Baeyens JP, Bauer JM, et al. Sarcopenia: European consensus on definition and diagnosis: Report of the European Working Group on Sarcopenia in Older People. Age Ageing. 2010;39:412-23.

21. Baumgartner RN, Koehler KM, Gallagher D, et al. Epidemiology of sarcopenia among the elderly in New Mexico. Am J Epidemiol. 1998;147:755-63.

22. Janssen I, Heymsfield SB, and Ross R. Low relative skeletal muscle mass (sarcopenia) in older persons is associated with functional impairment and physical disability. J Am Geriatr Soc. 2002;50:889-96.

23. Prado CM, Lieffers JR, McCargar LJ, et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. Lancet Oncol. 2008;9:629-35.

24. Fearon K, Strasser F, Anker SD, et al. Definition and classification of cancer cachexia: an international consensus. Lancet Oncol. 2011;12:489-95.

25. Evans WJ. Skeletal muscle loss: cachexia, sarcopenia, and inactivity. Am J Clin Nutr. 2010;91:1123S-1127S.

26. Evans WJ, Morley JE, Argiles J, et al. Cachexia: a new definition. Clin Nutr. 2008;27:793-9.

27. Bachmann J, Heiligensetzer M, Krakowski-Roosen H, et al. Cachexia worsens prognosis in patients with resectable pancreatic cancer. J Gastrointest Surg. 2008;12:1193-201.

28. Antoun S, Baracos VE, Birdsell L, et al. Low body mass index and sarcopenia associated with dose-limiting toxicity of sorafenib in patients with renal cell carcinoma. Ann Oncol. 2010;21:1594-8.

29. Prado CM, Baracos VE, McCargar LJ, et al. Sarcopenia as a determinant of chemotherapy toxicity and time to tumor progression in metastatic breast cancer patients receiving capecitabine treatment. Clin Cancer Res. 2009;15:2920-6.

30. Fearon KC. Cancer cachexia: developing multimodal therapy for a multidimensional problem. Eur J Cancer. 2008;44:1124-32.

31. Tisdale MJ. Mechanisms of cancer cachexia. Physiol Rev. 2009;89:381-410.

32. Acharyya S, Butchbach ME, Sahenk Z, et al. Dystrophin glycoprotein complex dysfunction: a regulatory link between muscular dystrophy and cancer cachexia. Cancer Cell. 2005;8:421-32.

33. Loberg RD, Bradley DA, Tomlins SA, et al. The lethal phenotype of cancer: the molecular basis of death due to malignancy. CA Cancer J Clin. 2007;57:225-41.

34. Prado CM, Birdsell LA, and Baracos VE. The emerging role of computerized tomography in assessing cancer cachexia. Curr Opin Support Palliat Care. 2009;3:269-75.

35. Moher D, Liberati A, Tetzlaff J, et al. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. Int J Surg. 2010;8:336-41.

36. Wells GA, Shea B, O'connell D, et al. *The Newcastle-Ottawa Scale (NOS) for assessing the quality of nonrandomised studies in meta-analyses*. [cited 2015 20 May]; Available from: http://www.ohri.ca/programs/clinical_epidemiology/oxford.asp.

37. Awad S, Tan BH, Cui H, et al. Marked changes in body composition following neoadjuvant chemotherapy for oesophagogastric cancer. Clin Nutr. 2012;31:74-77.

38. Yip C, Goh V, Davies A, et al. Assessment of sarcopenia and changes in body composition after neoadjuvant chemotherapy and associations with clinical outcomes in esophageal cancer. Eur Radiol. 2014;24:998-1005.

39. Voron T, Tselikas L, Pietrasz D, et al. Sarcopenia Impacts on Short- and Long-term Results of Hepatectomy for Hepatocellular Carcinoma. Ann Surg. 2014.

40. Itoh S, Shirabe K, Matsumoto Y, et al. Effect of body composition on outcomes after hepatic resection for hepatocellular carcinoma. Ann Surg Oncol. 2014;21:3063-3068.

41. Harimoto N, Shirabe K, Yamashita YI, et al. Sarcopenia as a predictor of prognosis in patients following hepatectomy for hepatocellular carcinoma. Br J Surg. 2013;100:1523-1530.

42. Lieffers JR, Bathe OF, Fassbender K, et al. Sarcopenia is associated with postoperative infection and delayed recovery from colorectal cancer resection surgery. Br J Cancer. 2012;107:931-936.

43. Reisinger KW, van Vugt JLA, Tegels JJW, et al. Functional compromise reflected by sarcopenia, frailty, and nutritional depletion predicts adverse postoperative outcome after colorectal cancer surgery. Ann Surg. 2014.

44. Van Vledder MG, Levolger S, Ayez N, et al. Body composition and outcome in patients undergoing resection of colorectal liver metastases. Br J Surg. 2012;99:550-557.

45. Peng PD, Van Vledder MG, Tsai S, et al. Sarcopenia negatively impacts short-term outcomes in patients undergoing hepatic resection for colorectal liver metastasis. HPB. 2011;13:439-446.

46. Sheetz KH, Zhao L, Holcombe SA, et al. Decreased core muscle size is associated with worse patient survival following esophagectomy for cancer. Dis Esophagus. 2013;26:716-722.

47. Peng P, Hyder O, Firoozmand A, et al. Impact of sarcopenia on outcomes following resection of pancreatic adenocarcinoma. J Gastrointest Surg. 2012;16:1478-86.

48. Sabel MS, Terjimanian M, Conlon ASC, et al. Analytic morphometric assessment of patients undergoing colectomy for colon cancer. J Surg Oncol. 2013;108:169-175.

49. Jung HW, Kim JW, Kim JY, et al. Effect of muscle mass on toxicity and survival in patients with colon cancer undergoing adjuvant chemotherapy. Supportive Care Cancer. 2014.

50. Rickles AS, Iannuzzi JC, Nieman DR, et al. Sarcopenia: Significant independent risk factor for poor survival following esophageal resection. Gastroenterology. 2013;144:S1124.

51. Hayashi T, Ozawa S, Kamachi K, et al. Impact of sarcopenia on long-term prognosis of patients with esophageal cancer after surgery. J Clin Oncol. 2014;32.

52. Hasselager R and Gogenur I. Core muscle size assessed by perioperative abdominal CT scan is related to mortality, postoperative complications, and hospitalization after major abdominal surgery: a systematic review. Langenbecks Arch Surg. 2014;399:287-95.

53. Shen W, Punyanitya M, Wang Z, et al. Total body skeletal muscle and adipose tissue volumes: estimation from a single abdominal cross-sectional image. J Appl Physiol (1985). 2004;97:2333-8.

54. Lang PO, Michel JP, and Zekry D. Frailty syndrome: a transitional state in a dynamic process. Gerontology. 2009;55:539-49.

55. Fearon KC, Glass DJ, and Guttridge DC. Cancer cachexia: mediators, signaling, and metabolic pathways. Cell Metab. 2012;16:153-66.

56. Argiles JM, Busquets S, Stemmler B, et al. Cancer cachexia: understanding the molecular basis. Nat Rev Cancer. 2014;14:754-62.

57. Zhou X, Wang JL, Lu J, et al. Reversal of cancer cachexia and muscle wasting by ActRIIB antagonism leads to prolonged survival. Cell. 2010;142:531-43.

58. Jameson GS, Hoff DDV, Weiss GJ, et al. Safety of the antimyostatin monoclonal antibody LY2495655 in healthy subjects and patients with advanced cancer. J Clin Oncol. 2012;30 (Suppl).

59. Dingemans AM, de Vos-Geelen J, Langen R, et al. Phase II drugs that are currently in development for the treatment of cachexia. Expert Opin Investig Drugs. 2014;23:1655-69.

60. Martin L, Birdsell L, Macdonald N, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol. 2013;31:1539-47.

APPENDICES

Appendix S1 Modified Newcastle–Ottawa Scale for cohort studies (short-term outcome). A study can be awarded a maximum of one star for each numbered item within the selection and outcome categories. A maximum of two stars can be given for comparability

Selection

1) Representativeness of the exposed cohort

a) truly representative of the average _____ (describe) in the community *

b) somewhat representative of the average _____ in the community *

c) selected group of users, e.g., nurses, volunteers

d) no description of the derivation of the cohort

- Star if demographics (age and gender) and indication of surgical resection are both clearly explained **and** reasonably representative; may be subjective

2) Selection of the non-exposed cohort

a) drawn from the same community as the exposed cohort *

b) drawn from a different source

c) no description of the derivation of the non-exposed cohort

- Star if sarcopenic patients were identified within the same cohort as non-sarcopenic patients

3) Ascertainment of exposure

a) secure record (e.g., surgical records) *

b) structured interview *

c) written self report

d) no description

- Star if computed tomography was used in the assessment of sarcopenia and presence of sarcopenia is clearly defined

4) Demonstration that outcome of interest was not present at start of study

a) yes *

b) no

- Star if any statement is given regarding absence of prior history associated with muscle wasting; may be subjective

Comparability

1) Comparability of cohorts on the basis of the design or analysis

a) study controls for _____ (select the most important factor) ¯

• Star if study controls patient gender

b) study controls for any additional factor ¯ (This criteria could be modified to indicate specific control for a second important factor)

• Star if study controls for patient age

Outcome

1) Assessment of outcome

a) independent blind assessment ¯

b) record linkage ¯

c) self report

d) no description

• Star if post-operative complications are clearly defined or scored on basis of a validated classification of surgical complications

2) Was follow-up long enough for outcomes to occur

a) yes (select an adequate follow-up period for outcome of interest) ¯

b) no

• Star if study reports a median follow-up exceeding 30 days

• Star if data regarding 30-day mortality is obtained through record linkage

3) Adequacy of follow-up of cohorts

a) complete follow up – all subjects accounted for ¯

b) subjects lost to follow-up unlikely to introduce bias – small number lost – over 80 % follow-up, or description provided of those lost) ¯

c) follow-up rate less than 80 % and no description of those lost

d) no statement

• Star if all subjects are accounted for.

• Star if more than 80 % described or if those lost to follow-up have been described.

Appendix S2 Modified Newcastle–Ottawa Scale for cohort studies (disease-free survival). A study can be awarded a maximum of one star for each numbered item within the selection and outcome categories. A maximum of two stars can be given for comparability

Selection

1) Representativeness of the exposed cohort

a) truly representative of the average _____ (describe) in the community *

b) somewhat representative of the average _____ in the community *

c) selected group of users, e.g., nurses, volunteers

d) no description of the derivation of the cohort

• Star if demographics (age and gender) and indication of surgical resection are both clearly explained **and** reasonably representative; may be subjective

2) Selection of the non-exposed cohort

a) drawn from the same community as the exposed cohort *

b) drawn from a different source

c) no description of the derivation of the non-exposed cohort

• Star if sarcopenic patients were identified within the same cohort as non-sarcopenic patients

3) Ascertainment of exposure

a) secure record (e.g., surgical records) *

b) structured interview *

c) written self report

d) no description

• Star if computed tomography was used in the assessment of sarcopenia and presence of sarcopenia is clearly defined

4) Demonstration that outcome of interest was not present at start of study

a) yes *

b) no

• Star if any statement is given regarding absence of prior history associated with muscle wasting; may be subjective

Comparability

1) Comparability of cohorts on the basis of the design or analysis

a) study controls for _____ (select the most important factor) *

- Star if study controls patient gender

b) study controls for any additional factor - (This criteria could be modified to indicate specific control for a second important factor)

- Star if study controls for patient age

Outcome

1) Assessment of outcome

a) independent blind assessment -

b) record linkage -

c) self report

d) no description

- Star if disease-free survival is obtained through patient records
- Star if disease-free survival is obtained through record linkage

2) Was follow-up long enough for outcomes to occur

a) yes (select an adequate follow-up period for outcome of interest) -

b) no

- Star if study reports a median follow-up exceeding 24 months

3) Adequacy of follow-up of cohorts

e) complete follow up – all subjects accounted for -

f) subjects lost to follow-up unlikely to introduce bias – small number lost – over 80 % follow-up, or description provided of those lost) -

g) follow-up rate less than 80 % and no description of those lost

h) no statement

- Star if all subjects are accounted for.
- Star if more than 80 % described or if those lost to follow-up have been described.

Appendix S3 Modified Newcastle–Ottawa Scale for cohort studies (overall survival). A study can be awarded a maximum of one star for each numbered item within the selection and outcome categories. A maximum of two stars can be given for comparability

Selection

1) Representativeness of the exposed cohort

a) truly representative of the average _____ (describe) in the community -

b) somewhat representative of the average _____ in the community -

c) selected group of users, e.g., nurses, volunteers

d) no description of the derivation of the cohort

- Star if demographics (age and gender) and indication of surgical resection are both clearly explained **and** reasonably representative; may be subjective

2) Selection of the non-exposed cohort

a) drawn from the same community as the exposed cohort -

b) drawn from a different source

c) no description of the derivation of the non-exposed cohort

- Star if sarcopenic patients were identified within the same cohort as non-sarcopenic patients

3) Ascertainment of exposure

a) secure record (e.g., surgical records) -

b) structured interview -

c) written self report

d) no description

- Star if computed tomography was used in the assessment of sarcopenia and presence of sarcopenia is clearly defined

4) Demonstration that outcome of interest was not present at start of study

a) yes -

b) no

- Star if any statement is given regarding absence of prior history associated with muscle wasting; may be subjective

Comparability

1) Comparability of cohorts on the basis of the design or analysis

a) study controls for _____ (select the most important factor) -

- Star if study controls patient gender

b) study controls for any additional factor - (This criteria could be modified to indicate specific control for a second important factor)

- Star if study controls for patient age

Outcome

1) Assessment of outcome

a) independent blind assessment -

b) record linkage -

c) self report

d) no description

- Star if overall survival is obtained through patient records
- Star if overall survival is obtained through record linkage

2) Was follow-up long enough for outcomes to occur

a) yes (select an adequate follow-up period for outcome of interest) -

b) no

- Star if study reports a median follow-up exceeding 24 months
- Star if study reports collection of survival data by record linkage using e.g., civil registrars at a time point exceeding at least 24 months

3) Adequacy of follow-up of cohorts

i) complete follow up – all subjects accounted for -

j) subjects lost to follow-up unlikely to introduce bias – small number lost – over 80 % follow-up, or description provided of those lost) -

k) follow-up rate less than 80 % and no description of those lost

l) no statement

- Star if all subjects are accounted for.
- Star if more than 80 % described or if those lost to follow-up have been described.

CHAPTER 6

Functional Compromise Reflected by Sarcopenia, Frailty, and Nutritional Depletion Predicts Adverse Postoperative Outcome after Colorectal Cancer Surgery

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ABSTRACT

Background: Functional compromise in elderly colorectal surgical patients is considered as a significant factor of impaired postoperative recovery. Therefore, the predictive value of pre-operative functional compromise assessment was investigated. Sarcopenia is a hallmark of functional compromise.

Methods: 310 consecutive patients who underwent oncologic colorectal surgery were enrolled in a prospective digital database. Sarcopenia was assessed using the L3 muscle index using Osirix® on pre-operative computed tomography (CT). Groningen Frailty Indicator (GFI) and Short Nutritional Assessment Questionnaire (SNAQ) scores were used to assess frailty and nutritional compromise. Predictors for anastomotic leakage, sepsis and mortality were analyzed by logistic regression analysis.

Results: Age was an independent predictor of mortality (p=0.04; odds ratio (OR), 1.17; 95% confidence interval (CI), 1.01 – 1.37). 30-day/in-hospital mortality rate in sarcopenic patients was 8.8% versus 0.7% in non-sarcopenic patients (p=0.001; OR 15.5; 95% CI, 2.00 – 120). Sarcopenia was not predictive for anastomotic leakage or sepsis. Combination of high SNAQ score, high GFI score and sarcopenia strongly predicted sepsis (p=0.001; OR, 25.1; 95% CI, 5.11 – 123), sensitivity, 46%; specificity, 97%; positive likelihood ratio, 13 (95% CI, 4.4 – 38); negative likelihood ratio, 0.57 (95% CI, 0.33 – 0.97).

Conclusions: Functional compromise in colorectal cancer surgery is associated with adverse postoperative outcome. Assessment of functional compromise by means of a nutritional questionnaire (SNAQ), a frailty questionnaire (GFI), and sarcopenia measurement (L3 muscle index) can accurately predict postoperative sepsis.

INTRODUCTION

With an increasingly aging population, the number of older cancer patients is concomitantly rising. Currently 50 percent of patients with colorectal cancer is aged 70 years or older [1]. Older patients are at increased risk for developing peri-operative complications and suffer from higher mortality rates [2]. Postoperative recovery plays a crucial role in cancer treatment with respect to survival, morbidity, and quality of life [3-5]. Hence, pre-operative assessment is important to identify patients at risk for developing postoperative complications. Widely accepted risk assessments are however considered to be subjective and imprecise and not focused on patients with cancer [6].

It is hypothesized that functional compromise in the older surgical patient is a significant predictor of postoperative complications [7, 8]. A widely investigated aspect of functional compromise is frailty. Frailty is a state of increased vulnerability towards stressors in older individuals, leading to an increased risk of developing adverse health outcomes [9]. However, the definition of frailty remains controversial and there is no consensus about the clinical use of frailty as a preoperative predictor for postoperative outcome in elderly patients [7]. Weight loss, low muscle strength, reduced physical activity, exhaustion, and decreased walking speed are elements of a physical definition of frailty [6, 8], whereas comorbidity, polypharmacy, physical functioning, nutritional and cognitive status, depression and social support are a more multidimensional tool to assess frailty [10-12]. In addition, sarcopenia is an important factor in functional compromise and depletion of skeletal muscle can occur in normal-weight, overweight and obese patients and does therefore not equal ordinary weight loss or cachexia [13, 14]. Malnutrition is another key element of functional compromise that is associated with poor clinical outcome [15]. In elderly patients, preoperative malnutrition is common, and worsens the condition of this already vulnerable population [16].

In this study we investigated whether functional compromise is associated with postoperative morbidity and mortality after colorectal surgery. The following parameters of functional compromise were evaluated: CT-based measurement of muscle mass, Groningen Frailty Index (GFI) [17] and Short Nutritional Assessment Questionnaire (SNAQ) [18].

METHODS

Patients

All consecutive patients who underwent oncologic colorectal surgery in a single non-academic center with a dedicated team of colorectal surgeons from January 2010 until May 2012 were enrolled in a prospective digital cohort and analyzed retrospectively. The only exclusion criterion was unavailability of a preoperative CT scan. Data in the database included characteristics of the primary tumor and oncologic staging, specifications of surgical treatment, chemotherapy, radiotherapy, and postoperative complications and included a preoperative CT-scan of the abdomen which was performed routinely for tumor staging.

Complications

The following complications were analyzed prospectively in detail: anastomotic leakage (surgically and/or radiologically) and/or intra-abdominal abscess diagnosed by CT examination or treated by percutaneous drainage or surgery, sepsis and septic shock (defined as two or more of the following criteria positive: (1) temperature >38°C or <36°C; (2) heart rate >90 bpm; (3) respiratory rate >20 breaths/min or $PaCO_2$ <32 mmHg; (4) white blood cell count >12x10^9/L, <4x10^9/L, or >10% immature (band) forms plus documented infection and hypotension despite adequate fluid resuscitation (in case of septic shock) [19]), and postoperative mortality (within 30 days postoperatively or within same period of hospital admission). The 30-day mortality for patients discharged within this period was verified by checking the municipal personal records database.

Pre-operative CT-based muscle measurements

All patients underwent a CT-scan of the abdomen as part of routine pre-operative assessment, which was delivered on CD-ROM or DVD. Measurements were performed using Osirix® Version 3.3 (32-bit; http://www.osirix-viewer.com). The cross-sectional skeletal muscle surface (cm^2) assessment of sarcopenia was performed at the level of the third lumbar vertebra (L3) on two consecutive transversal coupes on which both vertebral spines were visible [20]. The 'Grow Region (2D/3D Segmentation)' tool in the menu of the program facilitated to automatically select all skeletal muscle mass in one coupe. The distinction between different tissues is based on Hounsfield Units (HU). A threshold range of -30 HU to +110 HU was used for skeletal muscle. Muscles measured were: psoas, paraspinal, transverse abdominal, external oblique, internal oblique and rectus abdominis muscles (figure 1). Hand-adjustment of the selected areas was performed if necessary and the muscle area was calculated automatically [20]. The averages of the two measurements were used for calculations. Two investigators

independently measured all L3-muscle area surface parameters (J.v.V. and J.T.). A third investigator (K.R.) performed a random control measurement on 10% of the CT-scans. All investigators did not have specific skills in radiology.

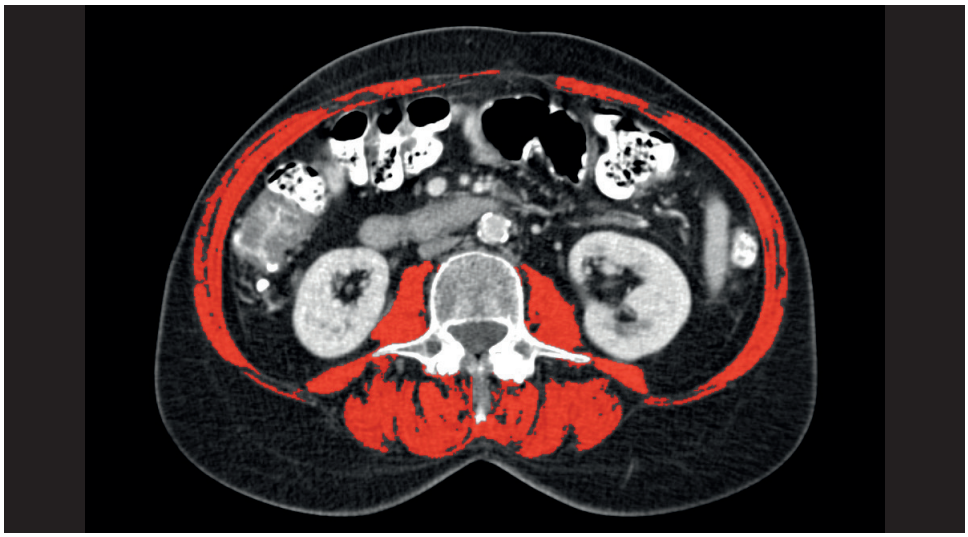

Figure 1. Computed tomography image at the third lumbar vertebral level. The following skeletal muscles are outlined in red: psoas, paraspinal, transverse abdominal, external oblique, internal oblique and rectus abdominis muscles. This female sarcopenic patient had an L3 muscle index of 34.3 cm^2/m^2.

Sarcopenia

The L3-muscle area surfaces were normalized for patient height to calculate the L3-muscle index and expressed in cm^2/m^2. The cut-off values used for sarcopenia were 52.4 cm^2/m^2 for men and 38.5 cm^2/m^2 for women, based on the method of Prado *et al.* [14].

Groningen Frailty Indicator (GFI)

The GFI has been developed as a simple screening instrument for frailty [17]. The GFI screens on physical, cognitive, social and emotional items (Appendix 1). The maximum score is 15 points. Patients scoring 5 or more points were considered frail [17]. A trained nurse routinely performed GFI-scores at pre-operative consultation or hospital admission in patients aged 70 or older.

Short Nutritional Assessment Questionnaire (SNAQ)

The SNAQ is a valid and reproducible tool to detect malnourished hospitalized patients without the need to calculate percentage weight loss or BMI [18]. The maximum score is 5 points (Appendix 2). Patients with a score of 3 points or more on the SNAQ were classified as malnourished (requiring nutritional support and supervision by a dietician) [18]. A trained nurse routinely performed SNAQ-scores at pre-operative consultation or hospital admission.

Statistical analysis

Frequencies are presented as absolute numbers and percentages. Continuous data are presented as mean (standard error of the mean [SEM]). Normality was tested using Kolmogorov-Smirnov. Differences between groups were analyzed with the Pearson chi-square test for dichotomous parameters. Odds ratios and 95% confidence intervals were calculated by logistic regression analysis. For the calculation of significant predictors of mortality and complications, univariate analyses with clinically relevant parameters were performed. Significant predictors ($p<0.05$) or predictors showing a trend towards significance ($0.05\leq p<0.20$) based on univariate analysis were entered into multivariable logistic regression analysis. Interactions between sarcopenia, GFI scores and SNAQ scores were tested. Functional compromise was defined as sarcopenia, high GFI score and high SNAQ score. Diagnostic accuracy of functional compromise assessment to predict postoperative complications was evaluated by calculating sensitivity, specificity, positive likelihood ratio (LR+), and negative likelihood ratio (LR-) using crosstabs. The interobserver agreement (J.v.V., J.T., K.R.) of the L3 muscle index assessment of sarcopenia was analyzed by Pearson's correlation and Cohen's kappa coefficient. Two-tailed p-values <0.05 were considered significant. All statistical analyses were performed using SPSS® 20.0 (SPSS Inc, Chicago, IL, USA).

RESULTS

Patients

340 patients were enrolled in the prospective database. In 30 patients, no staging CT scan was available, for which the main reason was emergency surgery. Therefore 310 patients undergoing both elective and acute colorectal resection were included in the analyses of the current study. The patient characteristics are listed in table 1. Of these 310 patients, 148 (47.7%) were identified as sarcopenic as defined by muscle index at L3 level; 90/155 (58.1%) males and 58/155 (37.4%) females. Mean L3 muscle indexes for males and females were 51.5 (0.65) cm^2/m^2 and 40.7 (0.53) cm^2/m^2, respectively. Of patients aged 70 or older, 41 (24.6%) were frail as defined by a GFI score of 5 or more. SNAQ scores were 3 points or higher in 36 (11%) of all patients.

Interobserver agreement of CT-based muscle measurement by Osirix®

Two independent investigators measured the L3 muscle areas of all patients, showing a strong and significant correlation (R^2=0.98; p<0.0001). Bland-Altman analysis produced 95% limits of agreement, -5.9% – 8.7%. The kappa (κ) of sarcopenia assessment by CT image analysis using Osirix® was 0.87 (95% confidence interval (CI), 0.82 – 0.93). The random control measurement on 10% of the CT-scans by a third investigator yielded excellent correlation (R^2=0.98; p<0.0001) and a kappa of 0.74 (95% CI, 0.46 – 1.00).

Mortality

Fourteen patients died within 30 days or during hospital admission (4.5%). Causes of mortality are listed in table 2. In univariate analyses sarcopenia (p=0.009), male gender (p=0.02), age (p=0.002), epidural analgesia (p=0.14), medical history of abdominal surgery (p=0.02), ASA classification (p=0.02), and stage 3-4 disease (p=0.14) were significant predictors of postoperative mortality or showed a trend towards significance. Multivariable logistic regression revealed male gender (OR, 49.29 (95% CI, 2.48 – 978.4); p=0.01), sarcopenia (OR, 43.30 (95% CI, 2.74 – 685.2); p=0.007), age (OR, 1.17; 95% CI, 1.01 – 1.37; p=0.04), and medical history of abdominal surgery (OR, 31.16 (3.09 – 313.9); p=0.004) being independent predictors of mortality. Logistic regression results are summarized in table 3a.

To evaluate the effect of functional compromise on mortality in more detail, interactions between SNAQ ≥3, GFI ≥5, and sarcopenia were studied. None of the interactions increased the predictive value of sarcopenia.

Table 1. patient characteristics

		Number of patients (%)	Mean (SEM)
Sex	Male	155 (50.0)	
	Female	155 (50.0)	
Age (years)			69 (0.6)[b]
	> 70	159 (51.3)	
BMI (kg/m^2)			26.3 (0.26)[b]
	> 25	182 (58.7)	
Length of hospital stay (days)			11 (0.6)[b]
ASA	I	17 (5.5)	
	II	219 (70.6)	
	III	62 (20.0)	
	IV	12 (3.9)	
Laparoscopy		33 (10.6)	
Primary anastomosis		249 (80.3)	
Tumor location	Colon	205 (66.1)	
	Rectum	105 (33.9)	
Complications	Anastomotic leakage and /or abscess	37 (14.9)	
	Sepsis	14 (4.5)	
	Death	14 (4.5)	
	Cause-related death	8 (2.6)	

Table 2. causes of mortality

	Cause	Postoperative day of mortality
Sarcopenic patients (n=13)		
1	Pneumonia, sepsis, cardiorespiratory failure	47
2	Sepsis of surgical site origin	5
3	Sepsis of surgical site origin	2
4	Sepsis of surgical site origin	2
5	Cardiorespiratory failure after anastomotic leakage	25
6	Pneumonia	30
7	Sepsis of surgical site origin	7
8	Pneumonia	3
9	Cardiorespiratory failure	8
10	Pneumonia	7
11	Unknown cause, probably pneumonia	35
12	Intraoperative presacral bleeding	0
13	Gastric hemorrhage	10
Non-sarcopenic patients (n=1)		
14	Anastomotic leakage	25

Table 3a. Logistic regression analysis for risk factors of mortality.

	Mortality rate	Univariate analysis		Multivariable analysis	
		Odds ratio	p-value	Odds ratio	p-value
Sarcopenia					
No	1/162	1		1	
Yes	13/148	15.50 (2.00-120.0)	0.009	43.30 (2.74-685.2)	0.007
GFI ≥5					
No	8/114	1			
Yes	3/39	0.68 (0.09-5.45)	0.72		
SNAQ ≥3					
No	8/185	1			
Yes	4/112	0.82 (0.24-2.79)	0.75		
Gender					
Female	2/155	1		1	
Male	12/155	6.42 (1.41-29.18)	0.02	49.29 (2.48-978.4)	0.01
Age		1.12 (1.04-1.20)	0.002	1.17 (1.01-1.37)	0.04
Epidural					
No	8/117	1			
Yes	6/193	0.44 (0.15-1.29)	0.14		
Previous abdominal surgery					
No	5/208	1		1	
Yes	9/102	3.93 (1.28-12.05)	0.02	31.16 (3.09-313.9)	0.004
ASA					
I	0/17	1	0.02		
II	5/219	∞ (0.00-∞)	1.00		
III	8/62	∞ (0.00-∞)	1.00		
IV	1/12	∞ (0.00-∞)	1.00		
Disease stage					
1-2	3/107	1			
3-4	9/122	2.76 (0.73-10.48)	0.14		
BMI >25 kg/m^2					
No	8/128	1			
Yes	6/182	0.51 (0.17-1.51)	0.23		
Smoking					
No	4/170	1			
Yes	8/140	2.12 (0.62-7.21)	0.23		
Diabetes					
No	11/252	1			
Yes	3/58	1.20 (0.32-4.43)	0.79		

Definition of mortality: 30-day mortality and/or in-hospital mortality
Values in parentheses are 95% confidence intervals

Anastomotic leakage

The incidence of anastomotic leakage and/or intra-abdominal abscess was 37 (14.9%) in patients with a primary anastomosis. Mortality within this group was 9.3%. Significant predictors or predictors showing a trend for anastomotic leakage and/or intra-abdominal abscess were sarcopenia (p=0.13), SNAQ ≥3 (p=0.08), medical history of abdominal surgery (p=0.06), surgery of the rectum (p=0.02), open surgery (p=0.10), stapled anastomosis (p=0.06), stage 3-4 disease (p=0.04), and need for blood transfusion (p<0.001). In multivariable analysis, stage 3-4 disease (OR, 3.68; 95% CI, 1.18 – 11.4; p=0.02) and need for blood transfusion (OR, 7.81; 95% CI, 2.81 – 21.8; p=0001) were independent predictors of anastomotic leakage and/or intra-abdominal abscess. Logistic regression results are summarized in table 3b.

To evaluate the effect of functional compromise on anastomotic leakage in more detail, interactions between SNAQ ≥3, GFI ≥5, and sarcopenia were studied. None of the interactions were significantly predictive of anastomotic leakage.

Sepsis

Fourteen patients (4.5%) developed sepsis. Mortality within this group was 36%. Univariate analysis demonstrated that GFI ≥5 (p=0.03), SNAQ ≥3 (p=0.003), age (p=0.03), epidural analgesia (p=0.14), and medical history of abdominal surgery (p=0.17) were significant predictors of sepsis or showed a trend towards significance. In multivariable logistic regression analysis, SNAQ ≥3 was independently predictive of postoperative sepsis (OR, 4.37; 95% CI, 1.07 – 17.9; p=0.04). Logistic regression results are summarized in table 3c.

To evaluate the effect of functional compromise on sepsis in more detail, interactions between SNAQ ≥3, GFI ≥5, and sarcopenia were studied. The interaction between SNAQ ≥3 and GFI ≥5 compared to SNAQ ≥3 alone increased the capability of predicting postoperative sepsis (OR, 13.1; 95% CI, 3.02 – 57.2; p=0.001). The interaction between SNAQ ≥3, GFI ≥5, and sarcopenia further increased the risk of sepsis (OR, 25.1; 95% CI, 5.11 – 123; p=0.001).

Table 3b. Logistic regression analysis for risk factors of anastomotic leakage.

	Anastomotic leakage rate	Univariate analysis		Multivariable analysis	
		Odds ratio	p-value	Odds ratio	p-value
Sarcopenia					
No	24/133	1			
Yes	13/116	0.57 (0.28-1.19)	0.13		
GFI ≥5					
No	12/92	1			
Yes	5/30	1.33 (0.42-4.15)	0.62		
SNAQ ≥3					
No	21/147	1			
Yes	14/91	2.33 (0.90-6.00)	0.08		
Gender					
Female	16/126	1			
Male	21/123	1.42 (0.70-2.86)	0.33		
Age		0.99 (0.96-1.02)	0.57		
Previous abdominal surgery					
No	20/168	1			
Yes	17/81	1.97 (0.97-4.00)	0.06		
Disease stage					
1-2	6/97	1		1	
3-4	17/109	2.80 (1.06-7.43)	0.04	3.68 (1.18-11.4)	0.02
Tumor location					
Colon	22/188	1			
Rectum	15/61	2.46 (1.18-5.12)	0.02		
Type of surgery					
Laparoscopy	1/29	1			
Laparotomy	36/220	5.48 (0.72-41.56)	0.10		
Anastomosis					
Manual	15/137	1			
Stapled	21/106	2.01 (0.98-4.12)	0.06		
Blood transfusion					
No	19/207	1		1	
Yes	18/42	7.42 (3.43-16.06)	<0.001	7.81 (2.81-21.8)	<0.001
Smoking					
No	17/133	1			
Yes	20/116	1.47 (0.70-3.08)	0.30		
Diabetes					
No	31/203	1			
Yes	6/46	0.83 (0.33-2.13)	0.70		

Anastomotic leakage and/or intra-abdominal abscess in 249 patients with primary anastomosis
Values in parentheses are 95% confidence intervals

Table 3c. Logistic regression analysis for risk factors of sepsis.

	Sepsis rate	Univariate analysis		Multivariable analysis	
		Odds ratio	p-value	Odds ratio	p-value
Sarcopenia					
No	6/162	1			
Yes	8/148	1.49 (0.50-4.39)	0.47		
GFI ≥5					
No	5/114	1			
Yes	6/39	3.96 (1.14-13.83)	0.03		
SNAQ ≥3					
No	6/185	1		1	
Yes	6/112	6.17 (1.84-20.64)	0.003	4.37 (1.07-17.9)	0.04
Gender					
Female	8/155	1			
Male	6/155	0.74 (0.25-2.19)	0.59		
Age		1.07 (1.01-1.14)	0.03		
Epidural					
No	8/117	1			
Yes	6/193	0.44 (0.15-1.29)	0.14		
Previous abdominal surgery					
No	7/208	1			
Yes	7/102	2.12 (0.72-6.20)	0.17		
Disease stage					
1-2	4/107	1			
3-4	4/122	0.87 (0.21-3.58)	0.85		
ASA					
I	0/17	1	0.95		
II	10/219	∞ (0.00-∞)	1.00		
III	4/62	∞ (0.00-∞)	1.00		
IV	0/12	1.00 (0.00-∞)	1.00		
Smoking					
No	7/170	1			
Yes	7/140	1.21 (0.40-3.70)	0.74		
Diabetes					
No	11/252	1			
Yes	3/58	1.62 (0.50-5.29)	0.42		

Values in parentheses are 95% confidence intervals

Diagnostic accuracy of SNAQ/GFI/sarcopenia score to predict sepsis

As a strong association of functional compromise (the combination of high SNAQ, high GFI, and sarcopenia) with sepsis was shown by the previous data, the clinical applicability of this functional compromise assessment needed further exploration. Therefore, the diagnostic accuracy of (pre-operative) functional compromise assessment to predict postoperative sepsis was investigated. A positive functional compromise test was defined as SNAQ ≥3, GFI ≥5, and sarcopenia (L3 muscle index <52.4 cm^2/m^2 for men, and <38.5 cm^2/m^2 for women). This resulted in sensitivity, 46%; specificity, 97%; LR+, 13 (95% CI, 4.4 – 38); LR-, 0.57 (95% CI, 0.33 – 0.97). Subgroup analysis of patients undergoing rectal surgery revealed a higher diagnostic accuracy; sensitivity, 67%; specificity, 98%; LR+, 29 (95% CI, 3.61 – 239); LR-, 0.34 (95% CI, 0.07 – 1.69).

DISCUSSION

The association of functional compromise and unfavorable postoperative outcome in colorectal surgery has been described before [8, 10]. Robinson and co-workers investigated frailty, an important aspect of functional compromise, using a scoring system based on 7 items of different domains and showed that frailty increased healthcare costs and 30-day readmission rates. However, postoperative complications were not specified [10]. Another study involving elderly colorectal surgery patients demonstrated a 4-fold increased risk for major postoperative complications when frailty was present [8]. The frailty assessment used was based on the criteria of Fried *et al.* [21], i.e. 3 or more items of the following: unintentional weight loss (10 lbs in past year), self-reported exhaustion, weakness (grip strength), slow walking speed, and low physical activity. The current study underlines and extends the abovementioned findings for several reasons. First, this is the largest cohort thus far addressing the influence of functional compromise including frailty on postoperative outcome in colorectal surgery. Second, the proposed functional compromise assessment is easy to perform as staging CT scans are routinely performed and SNAQ and GFI questionnaires can be completed in only few minutes time. In most Dutch hospitals, nutritional assessment and GFI assessments are part of standard care at hospital admission. Third, this study highlights metabolic and functional compromise to be associated with postoperative mortality in colorectal surgery.

As in the present study, elderly patients showed higher rates of short-term postoperative mortality and sepsis, pointing at a the role of sepsis causing postoperative death in these patients. Moreover, 9 out of 13 mortality cases in sarcopenic patients were caused by sepsis or pneumonia. In the context of major abdominal surgery, it could be assumed that sepsis may be the consequence of inadequate gut barrier function, especially in the cancer bearing host. Loss of intestinal barrier function is indeed correlated with sepsis [22-24]. In addition, it has been demonstrated that gut barrier dysfunction and endotoxemia develop concurrently with cachexia in a mouse model of colorectal neoplasia [25]. In the current study, low muscle mass strongly correlated with mortality, however the link with sepsis could not be made statistically. It could be hypothesized that sarcopenia reflects a state of prolonged catabolism impairing host immune function and leading to an inadequate response to inflammatory stimuli. A strong association with sepsis was found when muscle mass assessment was combined with SNAQ and GFI scores, indicating that muscle wasting as a reflection of the more comprehensive syndrome of functional compromise predicts postoperative morbidity. Although an elevated

inflammatory response has been observed in sarcopenic colorectal cancer patients [26], future studies should address the unraveled link between frailty, gut barrier function and development of sepsis.

A correlation between elevated serum markers of preoperative systemic inflammatory response and postoperative infectious complications and mortality has been established extensively [27, 28]. Specifically, the Glasgow Prognostic Score (GPS) comprising C-reactive protein (CRP) and albumin levels is an easily obtainable and accurate scoring system to predict postoperative morbidity and mortality in patients undergoing colorectal surgery [29]. The current study presents another point of view of potentially the same phenomenon, namely physical impairment of the cancer bearing host reflected as functional compromise and systemic inflammation. Cause and effect remain to be determined and in future studies, both SNAQ/GFI/sarcopenia score and GPS should be acquired, for combination of both scores may increase predictive accuracy.

Remarkably, anastomotic leakage and/or intra-abdominal abscess could not be predicted by sarcopenia, SNAQ or GFI scores, nor by their interactions. The predictive factors found in this study were stage 3-4 cancer, i.e. advanced disease and need for blood transfusion. This observation was in line with the results of the large prospective study of Boccola and colleagues [30]. The effect underlying the association between stage 3-4 disease and anastomotic leakage has not been investigated in our study, however a nutritional cause seems unlikely as a high SNAQ score was not independently predictive in multivariable analysis. An increased inflammatory state could be hypothesized in advanced cancer stage, but markers for inflammation were not included in our analyses.

Identifying surgical patients at risk for developing complications remains challenging. Therefore, an important clinical implication can be drawn from this study. The easy-to-perform functional compromise assessment presented here (SNAQ, GFI and sarcopenia) may be used to detect high-risk patients and to adapt treatment regimens accordingly, i.e. primary diverting ileostomy or colostomy. The preoperative period can be considered to improve functional parameters by nutritional support and physical exercise [31]. Exercise may increase physical functioning and overall quality of life in cancer patients [32, 33]. The best anabolic response is obtained when exercise is combined with nutritional support, such as essential amino acid ingestion [34, 35]. Physical exercise must be performed 2-3 days a week to increase muscle strength [36], therefore this should be supervised. Furthermore, many patients undergo neoadjuvant treatment which generates a timeframe for prehabilitation strategies. Finally, as the SNAQ/GFI/

sarcopenia score specifically predicts sepsis, intensive monitoring can be opted for in patients at risk. Future studies may elucidate whether these high-risk patients could benefit from selective decontamination of the digestive tract (SDD) [37].

The L3 muscle index measure is a widely available, objective and precise measurement for peri-operative assessment of lean body mass. In the current study, a very good interobserver variability was demonstrated by two investigators who had not received any specific radiological training. As mentioned before, an important remark should be made regarding the cut-off values of the L3 index to diagnose sarcopenia. As these values are based on obese cancer patients, they might not be applicable to all populations. Sarcopenia incidence by the definition in the present study was 48%, which is a rather high number in colorectal cancer patients who have not been reported to suffer from severe muscle wasting, potentially indicating an underestimated proportion of functional loss in this group. It is therefore highly desirable to define cut-off values based on healthy subjects and different cancer type populations. Nonetheless, the cut-off values described by Prado *et al.* are based on mortality prediction, which was supported in the current study. Another limitation is the retrospective nature of the study. However, all patients in the study cohort were included in a prospective database (Dutch Surgical Colorectal Audit) and only patients without an available staging CT scan were excluded. As a consequence, the probability of selection bias was negligible. Due to the retrospective nature of the study, DXA scans and other tests for sarcopenia were not available. However, CT-based muscle area measurement is a well-documented and readily obtainable alternative to detect sarcopenia [14]. The findings of the present study should nonetheless be confirmed in a second, prospective cohort including other tests for sarcopenia.

This study shows that functional compromise in colorectal cancer surgery is associated with adverse postoperative outcome. Moreover, assessment of functional compromise by means of a nutritional questionnaire (SNAQ), a frailty questionnaire (GFI), and CT-based sarcopenia measurement (L3 muscle index), i.e. the SNAQ/GFI/sarcopenia score can accurately predict postoperative sepsis.

REFERENCES

1. Jaarrapportage 2010: uitkomst van zorg registratie. Dutch Surgical Colorectal Audit [database on the Internet]2010.
2. Polanczyk CA, Marcantonio E, Goldman L, Rohde LE, Orav J, Mangione CM, et al. Impact of age on perioperative complications and length of stay in patients undergoing noncardiac surgery. Annals of internal medicine 2001; 134: 637-43.
3. Law WL, Choi HK, Lee YM, Ho JW. The impact of postoperative complications on long-term outcomes following curative resection for colorectal cancer. Ann Surg Oncol 2007; 14: 2559-66.
4. Tanaka K, Kumamoto T, Nojiri K, Matsuyama R, Takeda K, Endo I. Impact of Postoperative Morbidity on Long-Term Survival After Resection for Colorectal Liver Metastases. Ann Surg Oncol 2010.
5. Walker KG, Bell SW, Rickard MJ, Mehanna D, Dent OF, Chapuis PH, et al. Anastomotic leakage is predictive of diminished survival after potentially curative resection for colorectal cancer. Annals of surgery 2004; 240: 255-9.
6. Makary MA, Segev DL, Pronovost PJ, Syin D, Bandeen-Roche K, Patel P, et al. Frailty as a predictor of surgical outcomes in older patients. Journal of the American College of Surgeons 2010; 210: 901-8.
7. Partridge JS, Harari D, Dhesi JK. Frailty in the older surgical patient: a review. Age Ageing 2012; 41: 142-7.
8. Tan KY, Kawamura YJ, Tokomitsu A, Tang T. Assessment for frailty is useful for predicting morbidity in elderly patients undergoing colorectal cancer resection whose comorbidities are already optimized. American journal of surgery 2012; 204: 139-43.
9. Olde Damink SW, Jalan R, Deutz NE, Hayes PC, Soeters PB. Protein synthesis is severely diminished following a simulated upper GI bleed in patients with cirrhosis. J Hepatol 2008; 49: 726-31.
10. Robinson TN, Wu DS, Stiegmann GV, Moss M. Frailty predicts increased hospital and six-month healthcare cost following colorectal surgery in older adults. American journal of surgery 2011; 202: 511-4.
11. Hamrick MW, Ding KH, Pennington C, Chao YJ, Wu YD, Howard B, et al. Age-related loss of muscle mass and bone strength in mice is associated with a decline in physical activity and serum leptin. Bone 2006; 39: 845-53.
12. Bossola M, Muscaritoli M, Costelli P, Grieco G, Bonelli G, Pacelli F, et al. Increased muscle proteasome activity correlates with disease severity in gastric cancer patients. Annals of surgery 2003; 237: 384-9.

13. Baracos VE, Reiman T, Mourtzakis M, Gioulbasanis I, Antoun S. Body composition in patients with non-small cell lung cancer: a contemporary view of cancer cachexia with the use of computed tomography image analysis. The American journal of clinical nutrition 2010; 91: 1133S-7S.
14. Prado CM, Lieffers JR, McCargar LJ, Reiman T, Sawyer MB, Martin L, et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. Lancet Oncol 2008; 9: 629-35.
15. Covinsky KE, Martin GE, Beyth RJ, Justice AC, Sehgal AR, Landefeld CS. The relationship between clinical assessments of nutritional status and adverse outcomes in older hospitalized medical patients. Journal of the American Geriatrics Society 1999; 47: 532-8.
16. Mudge AM, Ross LJ, Young AM, Isenring EA, Banks MD. Helping understand nutritional gaps in the elderly (HUNGER): a prospective study of patient factors associated with inadequate nutritional intake in older medical inpatients. Clin Nutr 2011; 30: 320-5.
17. Steverink N, Slaets JPJ, Schuurmans H, Van Lis M. Measuring frailty: developing and testing the GFI (Groningen Frailty Indicator). Gerontologist 2001; 41: 236.
18. Kruizenga HM, Seidell JC, de Vet HC, Wierdsma NJ, van Bokhorst-de van der Schueren MA. Development and validation of a hospital screening tool for malnutrition: the short nutritional assessment questionnaire (SNAQ). Clin Nutr 2005; 24: 75-82.
19. Bone RC, Balk RA, Cerra FB, Dellinger RP, Fein AM, Knaus WA, et al. Definitions for sepsis and organ failure and guidelines for the use of innovative therapies in sepsis. The ACCP/SCCM Consensus Conference Committee. American College of Chest Physicians/Society of Critical Care Medicine. Chest 1992; 101: 1644-55.
20. Dello SA, Lodewick TM, van Dam RM, Reisinger KW, van den Broek MA, von Meyenfeldt MF, et al. Sarcopenia negatively affects preoperative total functional liver volume in patients undergoing liver resection. HPB (Oxford) 2013; 15: 165-9.
21. Fried LP, Tangen CM, Walston J, Newman AB, Hirsch C, Gottdiener J, et al. Frailty in older adults: evidence for a phenotype. J Gerontol A Biol Sci Med Sci 2001; 56: M146-56.
22. Deitch EA, Kemper AC, Specian RD, Berg RD. A study of the relationship among survival, gut-origin sepsis, and bacterial translocation in a model of systemic inflammation. J Trauma 1992; 32: 141-7.
23. Swank GM, Deitch EA. Role of the gut in multiple organ failure: bacterial translocation and permeability changes. World J Surg 1996; 20: 411-7.
24. Derikx JP, Poeze M, van Bijnen AA, Buurman WA, Heineman E. Evidence for intestinal and liver epithelial cell injury in the early phase of sepsis. Shock 2007; 28: 544-8.
25. Puppa MJ, White JP, Sato S, Cairns M, Baynes JW, Carson JA. Gut barrier dysfunction in the Apc(Min/+) mouse model of colon cancer cachexia. Biochim Biophys Acta 2011; 1812: 1601-6.

26. Richards CH, Roxburgh CS, MacMillan MT, Isswiasi S, Robertson EG, Guthrie GK, et al. The relationships between body composition and the systemic inflammatory response in patients with primary operable colorectal cancer. PLoS One 2012; 7: e41883.

27. Moyes LH, Leitch EF, McKee RF, Anderson JH, Horgan PG, McMillan DC. Preoperative systemic inflammation predicts postoperative infectious complications in patients undergoing curative resection for colorectal cancer. Br J Cancer 2009; 100: 1236-9.

28. Ishizuka M, Nagata H, Takagi K, Horie T, Kubota K. Inflammation-based prognostic score is a novel predictor of postoperative outcome in patients with colorectal cancer. Annals of surgery 2007; 246: 1047-51.

29. McMillan DC. The systemic inflammation-based Glasgow Prognostic Score: a decade of experience in patients with cancer. Cancer treatment reviews 2013; 39: 534-40.

30. Boccola MA, Buettner PG, Rozen WM, Siu SK, Stevenson AR, Stitz R, et al. Risk factors and outcomes for anastomotic leakage in colorectal surgery: a single-institution analysis of 1576 patients. World J Surg 2011; 35: 186-95.

31. Fearon KC. The 2011 ESPEN Arvid Wretlind lecture: cancer cachexia: the potential impact of translational research on patient-focused outcomes. Clin Nutr 2012; 31: 577-82.

32. Fong DY, Ho JW, Hui BP, Lee AM, Macfarlane DJ, Leung SS, et al. Physical activity for cancer survivors: meta-analysis of randomised controlled trials. Bmj 2012; 344: e70.

33. Mishra SI, Scherer RW, Snyder C, Geigle PM, Berlanstein DR, Topaloglu O. Exercise interventions on health-related quality of life for people with cancer during active treatment. The Cochrane database of systematic reviews 2012; 8: CD008465.

34. Breen L, Phillips SM. Skeletal muscle protein metabolism in the elderly: Interventions to counteract the 'anabolic resistance' of ageing. Nutr Metab (Lond) 2011; 8: 68.

35. Volpi E, Kobayashi H, Sheffield-Moore M, Mittendorfer B, Wolfe RR. Essential amino acids are primarily responsible for the amino acid stimulation of muscle protein anabolism in healthy elderly adults. The American journal of clinical nutrition 2003; 78: 250-8.

36. Oldervoll LM, Loge JH, Paltiel H, Asp MB, Vidvei U, Wiken AN, et al. The effect of a physical exercise program in palliative care: A phase II study. J Pain Symptom Manage 2006; 31: 421-30.

37. Sleijfer DT, Mulder NH, de Vries-Hospers HG, Fidler V, Nieweg HO, van der Waaij D, et al. Infection prevention in granulocytopenic patients by selective decontamination of the digestive tract. Eur J Cancer 1980; 16: 859-69.

APPENDICES

Appendix 1: items of Groningen Frailty Indicator (GFI) score [1]

Mobility

Is the patient able to carry out these tasks single handed without any help? (The use of help resources such as walking stick, walking frame, wheelchair, is considered independent)

1. Shopping
2. Walking around outside (around the house or to the neighbors)
3. Dressing and undressing
4. Going to the toilet

Physical Fitness

5. What mark does the patient give himself/herself for physical fitness? (scale 0 to 10)

Vision

6. Does the patient experience problems in daily life due to poor vision?

Hearing

7. Does the patient experience problems in daily life due to being hard of hearing?

Nourishment

8. During the last 6 months has the patient lost a lot of weight unwillingly? (3 kg in 1 month or 6 kg in 2 months)

Morbidity

9. Does the patient take 4 or more different types of medicine?

Cognition (Perception)

10. Does the patient have any complaints about his/her memory or is the patient known to have a dementia syndrome?

Psychosocial

11. Does the patient sometimes experience emptiness around him/her?
12. Does the patient sometimes miss people around him/her?
13. Does the patient sometimes feel abandoned?
14. Has the patient recently felt downhearted or sad?

15. Has the patient recently felt nervous or anxious?

Sum
Scoring:
Questions 1–4: Independent = 0; dependent = 1
Question 5: 0–6 = 1; 7–10 = 0
Questions 6–9: No = 0; yes = 1
Question 10: No and sometimes = 0; yes = 1
Questions 11–15: No = 0; sometimes and yes = 1

Appendix 2: items of Short Nutritional Assessment Questionnaire (SNAQ) score [2]

Item	**Score**
Did you lose weight unintentionally?	
More than 6 kg in the last 6 months	3
More than 3 kg in the last month	2
Did you experience a decreased appetite over the last month?	1
Did you use supplemental drinks or tube feeding over the last month?	1

References appendices

1. Steverink N, Slaets JPJ, Schuurmans H, Van Lis M. Measuring frailty: developing and testing the GFI (Groningen Frailty Indicator). Gerontologist 2001; 41: 236.
2. Kruizenga HM, Seidell JC, de Vet HC, Wierdsma NJ, van Bokhorst-de van der Schueren MA. Development and validation of a hospital screening tool for malnutrition: the short nutritional assessment questionnaire (SNAQ). Clin Nutr 2005; 24: 75-82.

CHAPTER 7

Skeletal Muscle Depletion is Associated with Severe Postoperative Complications in Patients Undergoing Cytoreductive Surgery with Hyperthermic Intraperitoneal Chemotherapy for Peritoneal Carcinomatosis of Colorectal Cancer

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ABSTRACT

Background: In patients undergoing colorectal cancer surgery, skeletal muscle depletion (sarcopenia) is associated with impaired postoperative recovery and decreased survival. The aim of this study was to determine whether skeletal muscle depletion could predict postoperative complications in patients undergoing CRS-HIPEC for peritoneal carcinomatosis of colorectal cancer.

Methods: All consecutive patients with an available preoperative CT scan who underwent CRS-HIPEC for peritoneal carcinomatosis of colorectal cancer in two centers were analyzed. Skeletal muscle mass was determined using the L3 muscle index on the preoperative CT scan. The cut-off values defined by Prado *et al.* were used to classify patients as sarcopenic or non-sarcopenic.

Results: In total, 90 out of 206 included patients (43.7%) were classified as sarcopenic. Sarcopenic patients underwent significantly more reoperations compared with non-sarcopenic patients (25.6% versus 12.1%, p=0.012). The mean L3 muscle index was significantly lower in patients who developed severe postoperative complications compared with patients without severe postoperative complications (43.3 cm^2/m^2 versus 47.0 cm^2/m^2, p=0.005). In a multivariable logistic regression model, L3 muscle index was the only parameter that was independently associated with the risk of severe postoperative complications (OR 0.93 [95% CI 0.87-0.99], p=0.018).

Conclusions: Skeletal muscle mass depletion, assessed using CT-based muscle mass measurements, is associated with an increased risk of severe postoperative complications in patients undergoing CRS-HIPEC for colorectal peritoneal carcinomatosis and could therefore be used in preoperative risk assessment.

INTRODUCTION

Peritoneal seeding of colorectal cancer (peritoneal carcinomatosis [PC]) occurs in approximately 7% of patients at primary surgery and in up to 19% of patients during follow-up [1]. CytoReductive Surgery (CRS) with Hyperthermic IntraPEritoneal Chemotherapy (HIPEC) improves outcome in patients with peritoneal carcinomatosis with 5-year survival rates up to 40% [2]. Nevertheless, this treatment option is associated with considerable rates of postoperative complications and mortality [3]. A recent study showed that the occurrence of severe postoperative complications in patients undergoing CRS-HIPEC for colorectal peritonitis carcinomatosis is associated with impaired overall and disease-free survival [4, 5].

Therefore, the development of preoperative risk assessment tools is of utmost importance, particularly in oncologic populations undergoing major surgery. Skeletal muscle depletion (sarcopenia), a key determinant of frailty and cancer cachexia, is associated with a complicated postoperative course and decreased survival in patients undergoing surgery for colorectal cancer [6-9]. However, the predictive value of skeletal muscle mass assessment in patients undergoing CRS-HIPEC has never been investigated. We hypothesized that preoperative skeletal muscle mass measurement could be an adequate method to predict severe postoperative complications in patients undergoing CRS-HIPEC for colorectal peritoneal carcinomatosis.

METHODS

Patients and data collection

All consecutive patients who underwent CRS-HIPEC for peritoneal carcinomatosis of colorectal cancer in the St Antonius Hospital, Nieuwegein, the Netherlands and the Catharina Hospital, Eindhoven, the Netherlands between April 2005 and December 2013 were enrolled into a prospectively maintained database and analyzed retrospectively. Only patients who underwent a complete CRS-HIPEC procedure were included in the current study. Patients without a computed tomography (CT)-scan suitable for skeletal muscle mass analysis or with unrecorded height were excluded. Data regarding patient characteristics (e.g., age, sex, medical history), disease status (number of regions affected with PC, primary tumor location, diagnosis of PC), treatment (operative time, estimated blood loss, number of bowel anastomosis, completeness of cytoreduction score [CCR]) and postoperative recovery (postoperative complications, mortality, length of hospital stay) were retrieved from the database. Tumor characteristics were obtained from postoperative pathology reports. Postoperative complications were classified according to the Clavien-Dindo Classification of Surgical Complications [10]. Severe postoperative complications were defined as Clavien-Dindo Classification ≥ 3 (i.e. the need for re-intervention, ICU-admission or death).

Surgical procedure

CRS-HIPEC was performed by two dedicated surgical teams as previously described [11]. In summary, tumor load was estimated after laparotomy using the simplified peritoneal cancer index [12] and all macroscopically suspected lesions were surgically removed (cytoreductive surgery). After cytoreductive surgery, the abdominal cavity was perfused with isotonic dialysis fluid with an inflow temperature of 42 °C. Mytomycin-C at a dose of 35 mg/m^2 was added to the perfusate using 28 French catheters when a stable temperature of 41 °C was reached. Intraperitoneal perfusion was continued for 90 minutes. Hereafter, the perfusion fluid was drained from the abdominal cavity, bowel continuity was restored and intra-abdominal drains were placed when appropriate. All patients were treated according to the Enhanced Recovery After Surgery (ERAS) principles [13].

Skeletal muscle mass measurements

Abdominal CT scans were routinely performed as part of the preoperative work-up. Measurements of the muscle surface were performed using OsiriX version 3.3 (32-bit; http://www.osirix-viewer.com) by a medically trained member of the research group (AV) according to previously conducted studies [9]. In summary, the cross-sectional

muscle surface (cm^2) was assessed at the level of the third lumbar vertebra (L3) on two consecutive transversal coupes on which both transverse processes were visible. Measurements included the psoas, paraspinal, transverse abdominal, external oblique, internal oblique and rectus abdominis muscles. The average of these two L3 muscle surface area measurements was normalized for patient height, resulting in the L3 muscle index (cm^2/m^2). Only preoperative abdominal CT scans were used. The cut-off values of 52.4 cm^2/m^2 for men and 38.5 cm^2/m^2 for women defined by Prado *et al.* were used to classify patients as sarcopenic (muscle depletion) or non-sarcopenic (no muscle depletion) [14].

Statistical analysis

Frequencies are presented as absolute numbers and percentages. Continuous data are presented as median with interquartile ranges (IQR). Normality of data was tested using the Kolmogorov-Smirnov test. Differences between groups were analyzed using the Pearson Chi-square test for dichotomous variables and Student t-test or Mann-Whitney U test for continuous variables, depending on normality. Two-tailed p-values <0.05 were considered statistically significant. Odds ratios (OR) with 95% confidence intervals (CI) were calculated using logistic regression analysis, which was performed to identify risk factors for the occurrence of severe postoperative complications. All significant variables resulting from the univariable logistic regression analysis were entered into a multivariable logistic regression analysis. Since the L3 muscle index greatly depends on sex, gender was also included in the multivariable logistic regression analysis. All statistical analyses were performed using IBM® SPSS® Statistics Version 21 (SPSS Inc., Chicago, IL, USA).

RESULTS

A total of 299 patients were enrolled in the two databases. An assessable abdominal CT scan was available in 206 (68.9%) patients. Since both hospitals are tertiary referral centers for HIPEC procedures, unavailability of a preoperative abdominal CT scan that was performed in the referring hospital was the most common reason for exclusion (n = 79). Other reasons for exclusion were the presence of an ostomy (n = 3) or tumor growth through the abdominal wall at L3-level (n = 1) leading to difficulties to accurately identify skeletal muscle tissue, artifacts (n = 2), missing of imaging at L3-level (n =1) or incomplete imaging of the abdominal wall muscles at the level of L3, for example due to obesity or deformities (n = 7).

In total, 90 (43.7%) patients were classified as sarcopenic. The time interval between abdominal CT and surgery did not significantly differ between the sarcopenic and non-sarcopenic groups (median 36 versus 28 days, p=0.281). Apart from BMI (median 23.5 kg/m^2 versus 26.4 kg/m^2, p<0.001) and regions affected with PC (mean 2.9 versus 3.4, p=0.034), baseline characteristics (table 1) and intra-operative outcomes (table 2) did not significantly differ between sarcopenic and non-sarcopenic patients. Sarcopenic patients underwent significantly more reoperations compared with non-sarcopenic patients (25.6% versus 12.1%, p=0.012) (table 2). Trends towards a significant difference in the occurrence of any postoperative complication (54.4% versus 41.4%, p=0.062), severe postoperative complications (33.3% versus 21.6%, p=0.058) and abdominal fistula (5.6% versus 0.9%, p=0.088) were observed.

The mean L3 muscle index was significantly lower in patients who developed severe postoperative complications (i.e. Clavien-Dindo Classification ≥ 3) compared with patients without severe postoperative complications (43.3 cm^2/m^2 versus 47.0 cm^2/m^2, p=0.005).

In univariable logistic regression analysis, an incremental L3 muscle index (OR 0.95 [95% CI 0.92-0.99], p=0.01), more affected regions with PC (OR 1.34 [95% CI 1.11-1.73], p=0.005), longer operative time (OR 1.31 [95% CI 1.07-1.59], p=0.008) and more estimated intra-operative blood loss (OR 1.49 [95% CI 1.06-2.08], p=0.021) were associated with the occurrence of severe postoperative complications (table 3). Together with gender, these factors were entered into the multivariable logistic regression model. An incremental L3 muscle index was the only parameter that remained independently associated with the risk of severe postoperative complications (OR 0.93 [95% CI 0.87-0.99], p=0.018).

Table 1. Patient and tumor characteristics.

	Muscle depletion n = 90 (43.7 %)	**No muscle depletion** n = 116 (56.3 %)	**p-value**
Gender			
Male	46 (51.1)	54 (46.6)	0.516
Female	44 (48.9)	62 (53.4)	
Age	62.1 (10.0)	60.3 (10.1)	0.206
ASA			
I	28 (32.6)	31 (29.2)	
II	56 (65.1)	71 (67.0)	0.774
III	2 (2.3)	4 (3.8)	
BMI	23.5 (21.3-25.3)	26.4 (24.1-30.0)	<0.001
Regions affected with PC	2.9 (1.5)	3.4 (1.4)	0.034
Diagnosis of initial PC			
Synchronous	53 (58.9)	64 (55.2)	0.593
Metachronous	37 (41.1)	52 (44.8)	
Tumor location			
Colon	76 (87.4)	100 (90.1)	0.544
Rectum	11 (12.6)	11 (9.9)	
Tumor stage			
1	0 (0.0)	1 (0.9)	
2	4 (4.5)	3 (2.7)	0.067
3	48 (53.9)	42 (37.5)	
4	37 (41.6)	66 (58.9)	
Lymph node stage			
0	24 (27.6)	34 (30.1)	
1	30 (34.5)	31 (27.4)	0.560
2	33 (37.9)	48 (42.5)	
Tumor differentiation			
Good	5 (6.3)	4 (4.6)	
Moderate	58 (73.4)	58 (66.7)	0.426
Poor	16 (20.3)	25 (28.7)	
Tumor histology			
Adenocarcinoma	66 (73.3)	79 (68.1)	
Mucinous carcinoma	21 (23.3)	32 (27.6)	0.713
Signet cell carcinoma	3 (3.3)	5 (4.3)	
Time interval CT - surgery (days)	36 (17-56)	28 (15-49)	0.281

Continuous parameters are presented as mean (SD) or median (IQR); categorical parameters are presented as number (%). ASA, American Society of Anesthesiologists classification; BMI, body mass index; PC, peritoneal carcinomatosis; CT, computed tomography of the abdomen

Table 2. Intra-operative and postoperative outcomes.

	Muscle depletion n = 90 (43.7 %)	**No muscle depletion n = 116 (56.3 %)**	**p-value**
Intra-operative outcomes			
Operative time (min)	400 (339.75-451.75)	398.5 (331.25-471)	0.991
Estimated blood loss (ml)	600 (300-1200)	700 (300-1500)	0.768
Cytoreduction CCR 0-1 CCR 2	 87 (96.7) 3 (3.3)	 113 (97.4) 3 (2.6)	0.752
Anastomosis (yes)	66 (74.2)	84 (72.4)	0.780
Stomy (yes)	41 (45.6)	47 (40.5)	0.468
Postoperative outcomes			
Any complication	49 (54.4)	48 (41.4)	0.062
Anastomotic leakage*	15 (22.7)	11 (13.1)	0.122
Intra-abdominal abscess	10 (11.1)	15 (12.9)	0.692
Ileus/gastroparesis	18 (20.0)	15 (12.9)	0.170
Abdominal fistula	5 (5.6)	1 (0.9)	0.088
Pneumonia	6 (6.7)	10 (8.6)	0.602
Pulmonary embolism	1 (1.1)	0 (0.0)	0.437
Urinary tract infection	6 (6.7)	9 (7.8)	0.765
Severe complication#	30 (33.3)	25 (21.6)	0.058
Reoperation	23 (25.6)	14 (12.1)	0.012
30-day/in hospital mortality	2 (2.2)	3 (2.6)	0.866
Length of stay (days)	11.0 (8-19.5)	11.5 (8-16)	0.646

Continuous parameters are presented as median (IQR); categorical parameters are presented as number (%). CCR, completeness of cytoreduction. * Data available from 150 patients with an anastomosis; # Clavien-Dindo Classification ≥ 3 (i.e. the need for re-intervention, ICU-admission or death)

Table 3. Logistic regression analysis for risk factors associated with severe postoperative complications (i.e. Clavien-Dindo Classification ≥ 3).

	Univariable OR (95% CI)	p-value	Multivariable OR (95% CI)	p-value
L3 muscle index (cm^2/m^2)	0.95 (0.91-0.99)	0.008	0.93 (0.87-0.99)	0.018
Gender				
Male	1	0.245	1	0.412
Female	1.45 (0.78-2.70)		0.66 (0.25-1.78)	
Age	1.01 (0.98-1.05)	0.439		
ASA				
I	1	0.364		
II	1.40 (0.68-2.90)	0.536		
III	1.77 (0.29-10.76)			
BMI	0.93 (0.86-1.01)	0.076		
Diagnosis of initial PC				
Synchronous	1	0.694		
Metachronous	1.13 (0.61-2.11)			
Tumor location				
Colon	1	0.576		
Rectum	0.74 (0.26-2.12)			
Tumor stage	0.83 (0.49-1.39)	0.467		
Lymph node stage	1.15 (0.79-1.69)	0.465		
Regions affected with PC	1.36 (1.08-1.70)	0.008	1.26 (0.96-1.66)	0.102
Operative time (hours)	1.32 (1.08-1.62)	0.006	1.19 (0.91-1.57)	0.210
Estimated blood loss (liters)	1.47 (1.05-2.05)	0.024	1.23 (0.84-1.80)	0.287
Anastomosis				
No	1	0.094		
Yes	1.93 (0.89-4.16)			

OR, Odds ratio; 95% CI, 95% confidence interval; ASA; American Society of Anesthesiologists classification; BMI, body mass index; PC, peritoneal carcinomatosis.

DISCUSSION

Our study supports the hypothesis that preoperative skeletal muscle mass measurements can be used to predict a complicated postoperative recovery in patients undergoing CRS-HIPEC for colorectal peritoneal carcinomatosis. Sarcopenic patients undergo significantly more reoperations compared with non-sarcopenic patients. Moreover, an incremental L3 muscle index is an independent predictor for a lower rate of severe postoperative complications.

The association between skeletal muscle depletion and the risk of severe postoperative complications has been established in various surgical oncology populations, among which patients undergoing hepatectomy for colorectal liver metastases [15]. Patients undergoing CRS-HIPEC may sustain severe postoperative complications and mortality rates up to 30% and 3% respectively [3]. Hence, preoperative risk assessment is of utmost importance to identify patients (un)fit for surgery and to assist patient and physician in the shared decision-making process. Moreover, the occurrence of severe postoperative complications is associated with impaired overall and disease-free survival [4, 5]. Currently available assessments are considered imprecise and not focused on oncologic patients [16]. For example, the widely used ASA (American Society of Anesthesiologists) classification is subjective with great inconsistency between anaesthesiologists [16, 17]. While an incremental L3 muscle index was protective for the occurrence of severe postoperative complications in our study, no association with ASA classification was found. Skeletal muscle mass assessment may more objectively reflect a patient's physiological reserves, which are essential to tolerate and rehabilitate from major interventions such as cytoreductive surgery. Therefore, it seems a valuable element in the development of preoperative risk assessment scores.

CT-based L3 muscle index measurement is an easily obtainable, objective, and precise method to assess skeletal muscle mass with a limited inter-observer variability [9]. Since all patients scheduled for CRS-HIPEC routinely undergo CT-imaging as part of the preoperative work-up, abdominal CT scans are widely available. The time interval between CT scan and surgery could potentially be used to optimize patients for surgery without causing a treatment delay. Optimizing treatment options could include nutritional support, physical exercise therapy and treatment of secondary anorexia causes (e.g., pain and nausea) and inflammation-related metabolic changes (e.g., by anti-inflammatory drugs) [18]. Considering surgical prehabilitation a fast effect of the anti-catabolic treatment would be desirable. Therefore, the development of drugs targeting pathways involved in skeletal muscle synthesis and breakdown is wanted. For

example, myostatin, inducing muscle depletion, and insulin-like growth factor (IGF)-1, stimulating protein synthesis, are potential therapeutic targets with promising results in animal models [19, 20].

Although the current study shows an association between an incremental L3 muscle index and decrease in the occurrence of severe postoperative complications, the clinical practicability would increase when cut-off values to define skeletal muscle depletion could be used instead of the continuous L3 muscle index. The currently used cut-off values, defined by Prado *et al.* and acknowledged in an international consensus meeting, are based on mortality prediction in obese patients with pulmonary and gastrointestinal cancer [14, 21]. These cut-off values were frequently used in previous surgical series [6, 9, 22], enabling comparison of outcome in various populations and studies. However, it remains questionable whether these cut-off values are generalizable for every patient population, particularly for a non-obese surgical population. In our study, the use of these cut-off values may have led to an overestimation of patients with low skeletal muscle mass, since the mean BMI was 25.6 kg/m^2 and only 15.5% of patients were classified as obese (BMI ≥ 30 kg/m^2). Nevertheless, the prevalence of skeletal muscle mass depletion in our study (43.7%) is comparable to two previous studies on skeletal muscle depletion in colorectal cancer surgery patients, reporting a prevalence of 38.9% and 47.7%, respectively [6, 9].

Interestingly, in patients with skeletal muscle depletion significantly fewer regions affected with peritoneal carcinomatosis and a trend towards a significantly lower tumor stage was observed than in those without. These findings suggest that skeletal muscle depletion is not solely determined by tumor load and that other factors may play a role. In a mouse model, interactions between tumor and its environing tissue, i.e. activation of cytokines and white blood cells, seemed to induce cancer cachexia [23]. Tumor and patient specific factors influencing skeletal muscle mass should be further investigated.

This study is limited by its retrospective nature, which may have led to selection bias. First, selection may have occurred in the referral of patients from other centers to our two HIPEC centers. Second, there is a selection of patients considered fit for surgery during preoperative consultation in our and the referring centers. This might have resulted in additional exclusion of sarcopenic patients, as they are likely to be unfit for surgery. Finally, during laparotomy it is decided to continue the procedure based on several criteria: significant tumor load in the mesenteries and extensive adhesions may be contraindication to precede the procedure. The current study cohort only consisted of patients who underwent a complete CRS-HIPEC procedure.

In conclusion, skeletal muscle mass depletion, assessed using CT-based muscle mass measurements, is associated with an increased rate of severe postoperative complications in patients undergoing CRS-HIPEC for colorectal peritoneal carcinomatosis and could be used in preoperative risk assessment. Future research should elucidate the etiology of skeletal muscle mass depletion and focus on developing effective treatment strategies to counteract its deleterious effects in patients undergoing surgery.

REFERENCES

1. Koppe MJ, Boerman OC, Oyen WJ, Bleichrodt RP. Peritoneal carcinomatosis of colorectal origin: incidence and current treatment strategies. Annals of surgery 2006; 243: 212-22.
2. Cao C, Yan TD, Black D, Morris DL. A systematic review and meta-analysis of cytoreductive surgery with perioperative intraperitoneal chemotherapy for peritoneal carcinomatosis of colorectal origin. Ann Surg Oncol 2009; 16: 2152-65.
3. Chua TC, Yan TD, Saxena A, Morris DL. Should the treatment of peritoneal carcinomatosis by cytoreductive surgery and hyperthermic intraperitoneal chemotherapy still be regarded as a highly morbid procedure?: a systematic review of morbidity and mortality. Annals of surgery 2009; 249: 900-7.
4. Baratti D, Kusamura S, Iusco D, Bonomi S, Grassi A, Virzi S, et al. Postoperative complications after cytoreductive surgery and hyperthermic intraperitoneal chemotherapy affect long-term outcome of patients with peritoneal metastases from colorectal cancer: a two-center study of 101 patients. Dis Colon Rectum 2014; 57: 858-68.
5. Simkens GA, van Oudheusden TR, Luyer MD, Nienhuijs SW, Nieuwenhuijzen GA, Rutten HJ, et al. Serious Postoperative Complications Affect Early Recurrence After Cytoreductive Surgery and HIPEC for Colorectal Peritoneal Carcinomatosis. Ann Surg Oncol 2014.
6. Lieffers JR, Bathe OF, Fassbender K, Winget M, Baracos VE. Sarcopenia is associated with postoperative infection and delayed recovery from colorectal cancer resection surgery. Br J Cancer 2012; 107: 931-6.
7. van Vledder MG, Levolger S, Ayez N, Verhoef C, Tran TC, IJzermans JN. Body composition and outcome in patients undergoing resection of colorectal liver metastases. Br J Surg 2012; 99: 550-7.
8. Miyamoto Y, Baba Y, Sakamoto Y, Ohuchi M, Tokunaga R, Kurashige J, et al. Sarcopenia is a Negative Prognostic Factor After Curative Resection of Colorectal Cancer. Annals of surgical oncology 2015.
9. Reisinger KW, van Vugt JL, Tegels JJ, Snijders C, Hulsewe KW, Hoofwijk AG, et al. Functional compromise reflected by sarcopenia, frailty, and nutritional depletion predicts adverse postoperative outcome after colorectal cancer surgery. Annals of surgery 2015; 261: 345-52.
10. Dindo D, Demartines N, Clavien PA. Classification of surgical complications: a new proposal with evaluation in a cohort of 6336 patients and results of a survey. Annals of surgery 2004; 240: 205-13.
11. Braam HJ, Boerma D, Wiezer MJ, van Ramshorst B. Hyperthermic intraperitoneal chemotherapy during primary tumour resection limits extent of bowel resection compared to two-stage treatment. Eur J Surg Oncol 2013; 39: 988-93.
12. Swellengrebel HA, Zoetmulder FA, Smeenk RM, Antonini N, Verwaal VJ. Quantitative intra-operative assessment of peritoneal carcinomatosis - a comparison of three prognostic tools. Eur J Surg Oncol 2009; 35: 1078-84.

13. Fearon KC, Ljungqvist O, Von Meyenfeldt M, Revhaug A, Dejong CH, Lassen K, et al. Enhanced recovery after surgery: a consensus review of clinical care for patients undergoing colonic resection. Clin Nutr 2005; 24: 466-77.
14. Prado CM, Lieffers JR, McCargar LJ, Reiman T, Sawyer MB, Martin L, et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. Lancet Oncol 2008; 9: 629-35.
15. Peng PD, van Vledder MG, Tsai S, de Jong MC, Makary M, Ng J, et al. Sarcopenia negatively impacts short-term outcomes in patients undergoing hepatic resection for colorectal liver metastasis. HPB (Oxford) 2011; 13: 439-46.
16. Makary MA, Segev DL, Pronovost PJ, Syin D, Bandeen-Roche K, Patel P, et al. Frailty as a predictor of surgical outcomes in older patients. Journal of the American College of Surgeons 2010; 210: 901-8.
17. Mak PH, Campbell RC, Irwin MG, American Society of A. The ASA Physical Status Classification: inter-observer consistency. American Society of Anesthesiologists. Anaesth Intensive Care 2002; 30: 633-40.
18. Fearon K, Arends J, Baracos V. Understanding the mechanisms and treatment options in cancer cachexia. Nature reviews Clinical oncology 2013; 10: 90-9.
19. Levolger S, Van Vledder MG, Huisman SA, De Bruin RWF, Ijzermans JNM. Inhibition of ALK 4/5 inhibits cancer cachexia associated muscle wasting. Eur Surg Res 2014; 52: 105.
20. Han HQ, Zhou X, Mitch WE, Goldberg AL. Myostatin/activin pathway antagonism: molecular basis and therapeutic potential. Int J Biochem Cell Biol 2013; 45: 2333-47.
21. Fearon K, Strasser F, Anker SD, Bosaeus I, Bruera E, Fainsinger RL, et al. Definition and classification of cancer cachexia: an international consensus. Lancet Oncol 2011; 12: 489-95.
22. Du Y, Karvellas CJ, Baracos V, Williams DC, Khadaroo RG, Acute C, et al. Sarcopenia is a predictor of outcomes in very elderly patients undergoing emergency surgery. Surgery 2014; 156: 521-7.
23. Chiba F, Soda K, Yamada S, Tokutake Y, Chohnan S, Konishi F, et al. The importance of tissue environment surrounding the tumor on the development of cancer cachexia. Int J Oncol 2014; 44: 177-86.

CHAPTER 8

Impact of Low Skeletal Muscle Mass and Density on Short and Long-Term Outcome after Resection of Stage I-III Colorectal Cancer: Results from a Prospective Multicenter Observational Study

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ABSTRACT

Background: Preoperative low skeletal muscle mass and density are associated with increased postoperative morbidity in patients undergoing curative colorectal cancer surgery. However, the long-term effects of low skeletal muscle mass and density remain uncertain.

Methods: Patients with stage I-III colorectal cancer undergoing surgery, enrolled in a prospective observational cohort study, were included. Skeletal muscle mass and density were measured on computed tomography examinations. Patients with high and low skeletal muscle mass and high and low skeletal muscle density, defined according to pre-established cut-offs, were compared regarding postoperative complications and mortality, disease-free survival (DFS), overall survival (OS), and cancer-specific survival (CSS).

Results: In total, 816 patients (53.9% males) with a median age of 70 (IQR 62-77) were included. About half of the patients had low skeletal muscle mass (50.4%) and density (64.1%). The rate of severe postoperative complications (Clavien-Dindo grade ≥3a) was significantly higher in patients with low versus high skeletal muscle and density (20.9% versus 13.6%, p=0.006; 20.0% versus 11.8%, p=0.003). Low skeletal muscle density was independently associated with severe postoperative complications (OR 1.89, 95% CI 1.11-3.23, p=0.020). The postoperative (i.e. 90-day) mortality rate was higher in patients with low skeletal muscle mass and density compared with patients with high skeletal muscle mass and density (3.6% versus 1.7%, p=0.091; and 3.4% versus 1.0%, p=0.038). No differences in DFS were observed. After adjustment for covariates such as age and Charlson Comorbidity Index, univariate differences in OS and CSS between patients with high and low skeletal muscle mass and density disappeared.

Conclusions: Low skeletal muscle mass and density are associated with short-term, but not long-term, outcome in patients undergoing curative colorectal cancer surgery. The preoperative period may be used to optimize patients.

INTRODUCTION

Colorectal cancer is one of the leading causes of cancer-related death with an estimated total cancer burden of 7% and a great impact on disability-adjusted life years worldwide [1,2]. Recently, there has been a rising interest in the impact of low skeletal muscle mass and density on short and long term outcome in cancer patients [3]. Skeletal muscle depletion may result from cancer, as part of the cancer-cachexia syndrome, and aging (i.e. sarcopenia, the involuntary age-related loss of skeletal muscle mass and strength) [4].

The impact of low skeletal muscle mass and density on postoperative outcome (i.e. postoperative complications and mortality) [2, 5-8] and chemotherapy toxicity [9-11] has frequently been described in colorectal cancer patients. Furthermore, low skeletal muscle mass and density are prognostic factors in patients undergoing surgery for colorectal metastases [12, 13] or chemotherapy for metastatic colorectal cancer [9, 14]. However, its effect on long -term outcome in patients with stage I-III colorectal cancer has been reported in only a few studies [15-17].

Therefore, the aim of the present study was to investigate the association between low skeletal muscle mass and density on both short and long-term outcome in patients undergoing colorectal cancer surgery with curative intent in a multi-center prospective study.

METHODS

Study design and patient selection

Patients were selected from the MATCH study, an ongoing prospective observational cohort study enrolling patients undergoing curative resection for primary colorectal cancer in seven centers in the region of Rotterdam, the Netherlands [18, 19]. The study was approved by the Institutional Review Board and all patients provided written informed consent (MEC-2007-088). All aspects listed in the STrengthening the Reporting of OBservational studies in Epidemiology (STROBE) guidelines were followed, and the paper was written accordingly [20].

All patients with stage I-III colorectal cancer, according to the 5th edition of the American Joint Cancer Committee staging manual, included in the first six years of the MATCH study (July 1st 2007 – July 1st 2013) were included in the current study. Exclusion criteria were unavailability of a preoperative computed tomography (CT) scan or unknown body mass index (BMI). Baseline characteristics (e.g., sex, age), tumor characteristics (e.g., tumor location and grade), surgical characteristics (e.g., type of resection, blood loss), and additional treatment (e.g., radio- or chemotherapy) were collected. Comorbidity was graded according to the American Society of Anesthesiologists (ASA) classification and the Charlson Comorbidity Index [21]. BMI was categorized in groups <20, 20-24.9, and ≥25 kg/m^2.

Outcome parameters

All postoperative complications occurring within 30 days after surgery, during hospital admission or during readmission within 30 days after discharge, were recorded. Severity was graded according to the Clavien-Dindo (CD) classification [22]. Severe complications were defined as CD grade ≥3a. The comprehensive complications index, integrating all complications including their severity in a scale from 0 (no complication) to 100 (death) [23], was calculated for each patient. Postoperative mortality was defined as mortality within 90 days postoperatively. Length of hospital stay (LOS) was calculated from the date of surgery to the date of discharge. Discharge status, i.e. home or other (rehabilitation, nursing home) was recorded.

Disease-free (DFS) and overall (OS) and survival were calculated from the date of surgery until recurrence or death, respectively. Survival status and cause of death were obtained from the Dutch Central Bureau of Statistics (CBS), which were used to

investigate differences in colorectal cancer-specific survival. Patients who were still alive on December 31, 2016 were censored. Patients who underwent a non-radical resection were excluded from the disease-free survival analysis.

Skeletal muscle mass and density measurements

Skeletal muscle mass and density were measured on contrast-enhanced CT scans, which were routinely performed as part of preoperative diagnosis and work-up. Only preoperative CT examinations were included. In rectal cancer patients, only CT examinations performed after neoadjuvant therapy were included.

As previously described [13], the total cross-sectional muscle area (cm^2) at the level of the third lumbar vertebra (L3) was selected and adjusted for patients' height using a validated software package [24]. This resulted in the skeletal muscle index (SMI; cm^2/m^2). The mean muscle attenuation (in Hounsfield Units [HU]) of the cross-sectional muscle area was noted as a measure of skeletal muscle density.

The cutoff values used to define low skeletal muscle mass were 41 cm^2/m^2 for women, 43 cm^2/m^2 for men with a BMI <25 kg/m^2 and 53 cm^2/m^2 for men with a BMI ≥25 kg/m^2 as described by Martin *et al* [25]. Low skeletal muscle density was defined as HU <41 for patients with a BMI <25 kg/m^2 and HU <33 for patients with a BMI ≥25 kg/m^2 [25].

Statistical analysis

Frequencies are presented in absolute numbers and percentages. Continuous data are presented as median with the interquartile range (IQR). Differences between groups were tested using the Chi-squared and Mann-Whitney U tests where appropriate. Kaplan Meier estimates and Cox regression analysis were used for the survival analysis. Factors with a significance level of $p<0.1$ in the univariate analysis were selected for a multivariable analysis with the factor of interest (low skeletal muscle mass and/or density). Univariate and multivariable logistic regression analysis was performed to investigate the association between low skeletal muscle mass and density and the occurrence of severe complications. All analyses were performed using SPSS for Windows version 22 (IBM Corp., Armonk, NY, USA). Two-sided p-values <0.05 were considered to be statistically significant.

RESULTS

Patient characteristics

A total of 981 patients with stage I-III colorectal cancer were included in the MATCH-study in the period July 1, 2007 to July 1, 2013. No preoperative CT examination was available in 157 patient records and BMI could not be retrieved from 8 patient charts, leaving a total sample size of 816 patients. The median time between CT and surgery was 33 (IQR 22-47) days. Baseline characteristics of included and excluded patients did not differ significantly, besides type of surgery (open surgery 41.9% in included patients versus 52.1% in excluded patients, p=0.032), number of patients with a stoma (72.2% versus 59.4%, p=0.001), and number of colon tumors (71.1% versus 61.2%, p=0.012).

All patient characteristics are listed in table 1 and 2. Just over half of the patients (50.4%) were considered to have low skeletal muscle mass before surgery, which was more often observed in colon cancer rather than rectal cancer patients (52.9% vs 44.5%, p=0.029). In the low skeletal muscle mass group, 44.9% of the patients were male compared to 63.1% in the group with high skeletal muscle mass (p<0.001). Furthermore, median age was significantly higher in patients with low skeletal muscle mass, whereas BMI was significantly lower (both p<0.001). Although the median Charlson Comorbidity Index was not significantly different for the two groups, ASA classification was significantly higher in patients with low skeletal muscle mass (p=0.044).

Similar differences were observed between the low and high skeletal muscle density group for sex, age, BMI, ASA classification, tumor location, and adjuvant therapy. Interestingly, the CCI was significantly higher in the low skeletal muscle density group compared with the high skeletal muscle density group (p<0.001). Patients in the low skeletal muscle density group had a lower tumor stage compared with the high density group (p=0.002). However, the median age of stage III patients was significantly lower compared with stage I and II (66.5 versus 70 and 72, respectively, p<0.001). Patients in the low skeletal muscle density group less often received neoadjuvant chemo- and/or radiotherapy (p=0.002).

Table 1. Baseline characteristics stratified for high and low skeletal muscle mass.

	High skeletal muscle mass n = 404 (49.5%)	**Low skeletal muscle mass n = 412 (50.5%)**	**p-value**
Sex (male)	255 (63.1)	185 (44.9)	<0.001
Age	66 (59-74)	73 (65-79)	<0.001
BMI			
<20	2 (0.5)	25 (6.1)	<0.001
20-24.9	171 (42.3)	152 (36.9)	
≥25.0	231 (57.2)	235 (57.0)	
Charlson comorbidity index	0 (0-2)	0 (0-1)	0.831
ASA classification			
I	80 (19.8)	50 (12.1)	0.044
II	250 (61.9)	278 (67.5)	
III	65 (16.1)	72 (17.5)	
IV	6 (1.5)	7 (1.7)	
V	0 (0.0)	1 (0.2)	
CEA	3.4 (1.9-7.1)	3.3 (2.0-8.3)	0.251
Type of surgery			
Open	161 (39.9)	180 (43.7)	0.282
Laparoscopic	197 (48.8)	198 (48.1)	
Conversion	44 (10.9)	33 (8.0)	
Stoma	122 (30.2)	104 (25.2)	0.104
Blood loss (ml)	150 (30-350)	100 (30-300)	0.570
Duration of surgery (minutes)	150 (114-195)	150 (110-194)	0.837
Surgical margins			
Positive	8 (2.0)	15 (3.6)	0.155
Negative	393 (97.3)	396 (96.1)	
Tumor stage			
I	131 (32.4)	124 (30.1)	0.080
II	130 (32.2)	163 (39.6)	
III	143 (35.4)	125 (30.3)	
Tumor location			
Colon	273 (67.6)	307 (74.5)	0.029
Rectum	131 (32.4)	105 (25.5)	
Tumor grade			
Good	106 (26.2)	113 (27.4)	0.979
Moderate	239 (59.2)	239 (58.0)	
Poor	43 (10.6)	43 (10.4)	
Other	8 (2.0)	17 (4.1)	
Angioinvasive growth	35 (8.7)	35 (8.5)	0.910
Neoadjuvant therapy	111 (27.5)	97 (23.5)	0.205
Adjuvant therapy			
Systemic	91 (22.5)	67 (16.3)	0.043
Radiotherapy	1 (0.2)	0 (0.0)	

Table 2. Baseline characteristics stratified for high and low skeletal muscle density.

	High skeletal muscle density n = 288 (35.3%)	**Low skeletal muscle density n = 524 (64.1%)**	**p-value**
Sex (male)	189 (65.6)	249 (47.5)	<0.001
Age	63 (56-69)	74 (66-80)	<0.001
BMI			
<20	9 (3.1)	18 (3.4)	0.034
20-24.9	97 (33.7)	224 (42.7)	
≥25.0	182 (63.2)	282 (53.8)	
Charlson comorbidity index	0 (0-1)	1 (0-2)	<0.001
ASA classification			
I	76 (26.4)	54 (10.3)	<0.001
II	183 (63.5)	341 (65.1)	
III	25 (8.7)	112 (21.4)	
IV	1 (0.3)	12 (2.3)	
V	0 (0.0)	1 (0.2)	
CEA	3.1 (2.0-7.9)	3.5 (2.0-7.4)	0.433
Type of surgery			
Open	107 (37.2)	232 (44.3)	0.119
Laparoscopic	153 (53.1)	241 (46.0)	
Conversion	26 (9.0)	50 (9.5)	
Stoma	88 (30.6)	137 (26.1)	0.170
Blood loss (ml)	150 (20-400)	150 (40-300)	0.488
Duration of surgery (minutes)	158 (120-196)	150 (110-194)	0.130
Surgical margins			
Positive	8 (2.8)	15 (2.9)	0.970
Negative	276 (95.8)	509 (97.1)	
Tumor stage			
I	89 (30.9)	164 (31.3)	0.002
II	84 (29.2)	207 (39.5)	
III	115 (39.9)	153 (29.2)	
Tumor location			
Colon	190 (66.0)	387 (73.9)	0.018
Rectum	98 (34.0)	137 (26.1)	
Tumor grade			
Good	86 (29.9)	133 (25.4)	0.354
Moderate	161 (55.9)	313 (59.7)	
Poor	28 (9.7)	59 (11.3)	
Other	8 (2.8)	9 (1.7)	
Angioinvasive growth	23 (8.0)	47 (9.0)	0.727
Neoadjuvant therapy	92 (31.9)	115 (21.9)	0.002
Adjuvant therapy			
Systemic	76 (26.4)	82 (15.6)	0.001
Radiotherapy	0 (0.0)	1 (0.2)	

Postoperative complications and mortality

The prevalence of (severe) postoperative complications, postoperative mortality and differences in length of stay and discharge status are shown in table 3. A significantly higher proportion of patients with low skeletal muscle mass (20.9% versus 13.6%, p=0.006) and density (20.0% versus 11.8%, p=0.003) experienced at least one severe postoperative complication (CD grade ≥3a). The Comprehensive Complication Index was significantly higher in patients with low skeletal muscle density compared with patients with high skeletal muscle density (p<0.001), whereas a non-significant difference was found between patients with low and high skeletal muscle mass (p=0.121). In multivariable analysis, low skeletal muscle mass was independently associated with the occurrence of severe postoperative complications (OR 1.89, 95% CI 1.11-3.23, p=0.020), whereas a non-significant association was found for low skeletal muscle density (OR 1.84, 95% CI 0.99-3.41, p=0.053) (table 4).

LOS was significantly longer in patients with low compared with high skeletal muscle mass (8 [IQR 6-13] versus 7 [IQR 5-10] days, p=0.035) and in patients with low compared with high skeletal muscle density (8 [IQR 6-14] versus 7 [IQR 5-10] days, p<0.001). Patients with low skeletal muscle mass and density were significantly more often discharged to a place other than home (p=0.035 and p<0.001, respectively).

The postoperative (i.e. 90-day) mortality rate was higher in patients with low skeletal muscle mass and density compared with patients with high skeletal muscle mass and density (3.6% versus 1.7%, p=0.091; and 3.4% versus 1.0%, p=0.038, respectively). Due to the small number of events, a multivariable analysis was not performed for 90-day mortality.

Disease-free survival

Neither low skeletal muscle mass (HR 1.17, 95% CI 0.84-1.62, p=0.356) nor low skeletal muscle density (HR 1.17, 95% CI 0.83-1.64, p=0.386) were associated with disease-free survival (table 5). The 1-year, 3-year, and 5-year DFS rates did not significantly differ between patients with low and high skeletal muscle mass (89.8%, 81.1%, and 77.1% versus 92.4%, 84.4%, and 81.1%; p=0.233, p=0.239, and p=0.207, respectively), nor between patients with low and high skeletal muscle density (91.0%, 82.7%, and 78.1% versus 91.4%, 83.2%, and 80.9%; p=0.859, p=0.855, and p=0.557, respectively). Therefore, no multivariable analysis was performed.

Table 3. Postoperative complications and recovery.

	High skeletal muscle mass	Low skeletal muscle mass	p-value	High skeletal muscle density	Low skeletal muscle density	p-value
Complications (overall)	187 (46.3)	197 (47.8)	0.662	111 (38.5)	271 (51.7)	<0.001
Severe complications (CD ≥ 3)	55 (13.6)	86 (20.9)	0.006	34 (11.8)	105 (20.0)	0.003
Comprehensive complication index	0 (0-21)	0 (0-21)	0.135	0 (0-21)	9 (0-21)	<0.001
LOS (days)	7 (5-11)	8 (6-13)	0.022	7 (5-10)	8 (6-14)	<0.001
Discharge to home	359 (95.7)	364 (91.5)	0.035	268 (98.2)	435 (91.0)	0.001

Table 4. Multivariable logistic regression model for severe postoperative complications (i.e. CD ≥ 3).

	Adjusted OR (95% CI)	p-value
Skeletal muscle mass		
High	1	0.018
Low	1.91 (1.12-3.25)	
Low skeletal muscle density		
High	1	0.045
Low	1.87 (1.01-3.46)	
Age, years	1.01 (0.98-1.03)	0.731
Charlson comorbidity index	1.15 (0.99-1.33)	0.063
Type of surgery		
Open	1	0.131
Laparoscopic	0.66 (0.38-1.13)	0.756
Conversion	1.13 (0.53-2.42)	
Stoma		
No	1	0.465
Yes	1.30 (0.64-2.63)	
Blood loss (100 ml)	1.06 (1.02-1.10)	0.002
Tumor location		
Colon	1	0.859
Rectum	0.89 (0.23-3.42)	
Neoadjuvant therapy		
No	1	0.544
Yes	1.54 (0.38-6.29)	

Table 5. Cox proportional hazards analysis for disease-free survival.

	HR (95% CI)	p-value
Skeletal muscle mass		
High	1	0.356
Low	1.17 (0.84-1.62)	
Low skeletal muscle density		
High	1	0.386
Low	1.17 (0.83-1.64)	
Sex		
Female	1	0.694
Male	1.07 (0.77-1.48)	
Age	1.02 (1.00-1.04)	0.027
BMI		
<20	1	0.780
20-24.9	0.87 (0.31-2.40)	0.765
≥25.0	1.17 (0.43-3.17)	
Charlson comorbidity index	1.09 (0.98-1.21)	0.103
ASA classification		
I	1	0.302
II	1.28 (0.80-2.03)	0.900
III	1.04 (0.56-1.93)	0.975
IV	0.98 (0.23-4.16)	
V	-	
CEA	1.00 (1.00-1.01)	0.154
Type of surgery		
Open	1	0.212
Laparoscopic	0.80 (0.57-1.13)	0.710
Conversion	1.11 (0.64-1.92)	
Stoma		
No	1	<0.001
Yes	1.94 (1.39-2.70)	
Blood loss (100 ml)	1.02 (1.00-1.05)	0.114
Duration of surgery (hours)	1.02 (0.87-1.20)	0.802
Tumor stage		
I	1	0.619
II	1.13 (0.70-1.84)	<0.001
III	2.88 (1.88-4.41)	
Tumor location		
Colon	1	0.001
Rectum	1.72 (1.24-2.40)	
Tumor grade		
Good	1	0.484
Moderate	0.87 (0.59-1.29)	0.141
Poor	1.50 (0.87-2.56)	0.152
Other	1.98 (0.78-5.04)	

	HR (95% CI)	p-value
Angioinvasive growth		
No	1	0.013
Yes	1.85 (1.14-3.00)	
Neoadjuvant therapy		
No	1	0.001
Yes	1.77 (1.27-2.48)	
Adjuvant therapy		
No	1	0.643
Yes	0.91 (0.60-1.37)	
Comprehensive complication index	1.01 (1.00-1.02)	0.058

Patients with positive surgical margins were excluded.

Overall-survival and cancer-specific survival

After a median follow-up of 76.5 months, 236 of the 816 (28.9%) patients had died. Eight patients (1.0%) were lost to follow-up. The median 1-year, 3-year, and 5-year OS estimates were 93.1%, 82.3%, and 73.3% in patients with low skeletal muscle mass compared with 95.3%, 87.8%, and 78.8% in patients with high skeletal muscle mass (p=0.193, p=0.029, and p=0.414, respectively), and 92.6%, 82.2%, and 72.3% in patients with low skeletal muscle density compared with 97.2%, 90.3%, and 82.9% in patients with high skeletal muscle density (p=0.007, p=0.002, and p=0.089, respectively) (figure 1).

In univariate analysis, both low skeletal muscle mass (HR 1.35, 95% CI 1.10-1.74, p=0.024) and low skeletal muscle density (HR 1.86, 95% CI 1.38-2.50, p<0.001) were associated with decreased OS. However, no significant association was found in multivariable analysis after adjustment for covariates such as age, Charlson Comorbidity Index, tumor stage, and the Comprehensive Complication Index (table 6).

A significant difference in cancer-specific survival was observed between patients with low and high skeletal muscle density (p=0.046), whereas no significant difference was observed between patients with low and highs skeletal muscle mass (p=0.235) (figure 2). However, in multivariable analysis no significant associations were found.

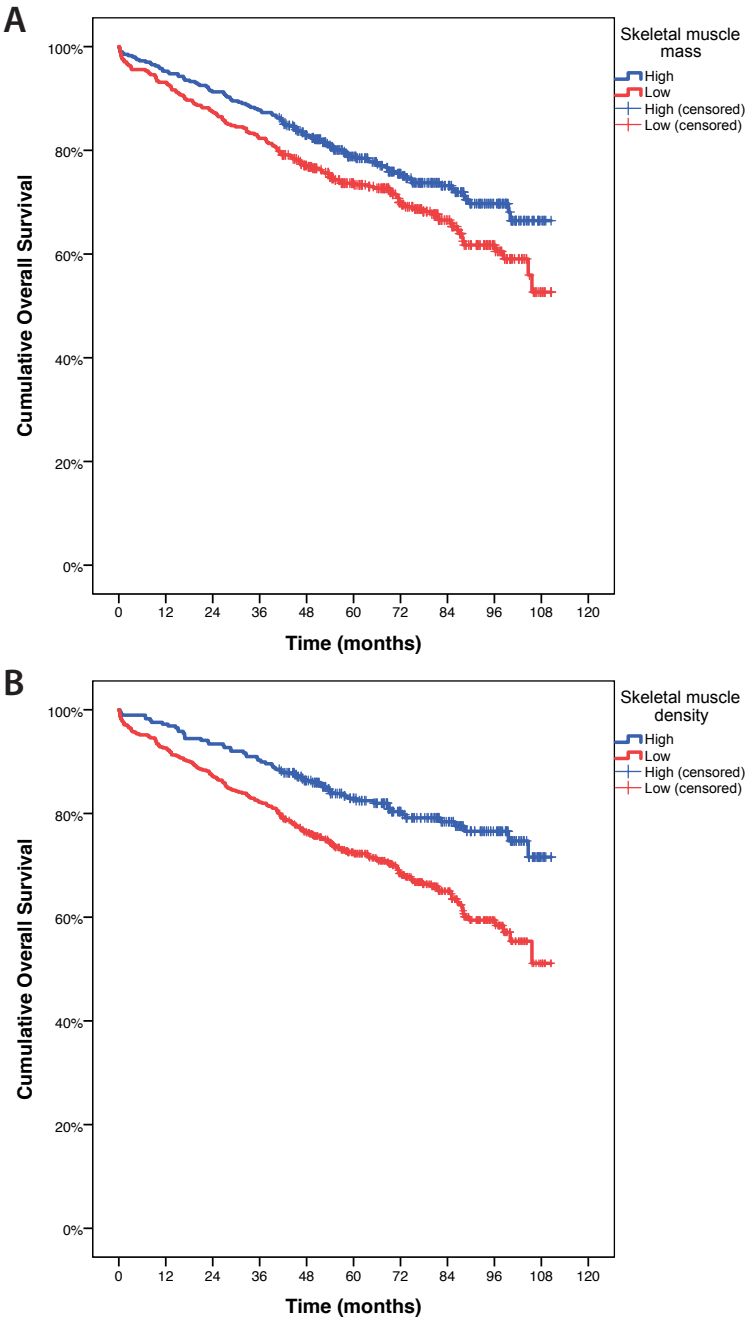

Figure 1. A. Kaplan-Meier curve for overall survival for patients with low skeletal muscle mass and high skeletal muscle mass (p=0.023). B. Kaplan-Meier curve for overall survival for patients with low skeletal muscle density and high skeletal muscle density (p<0.001).

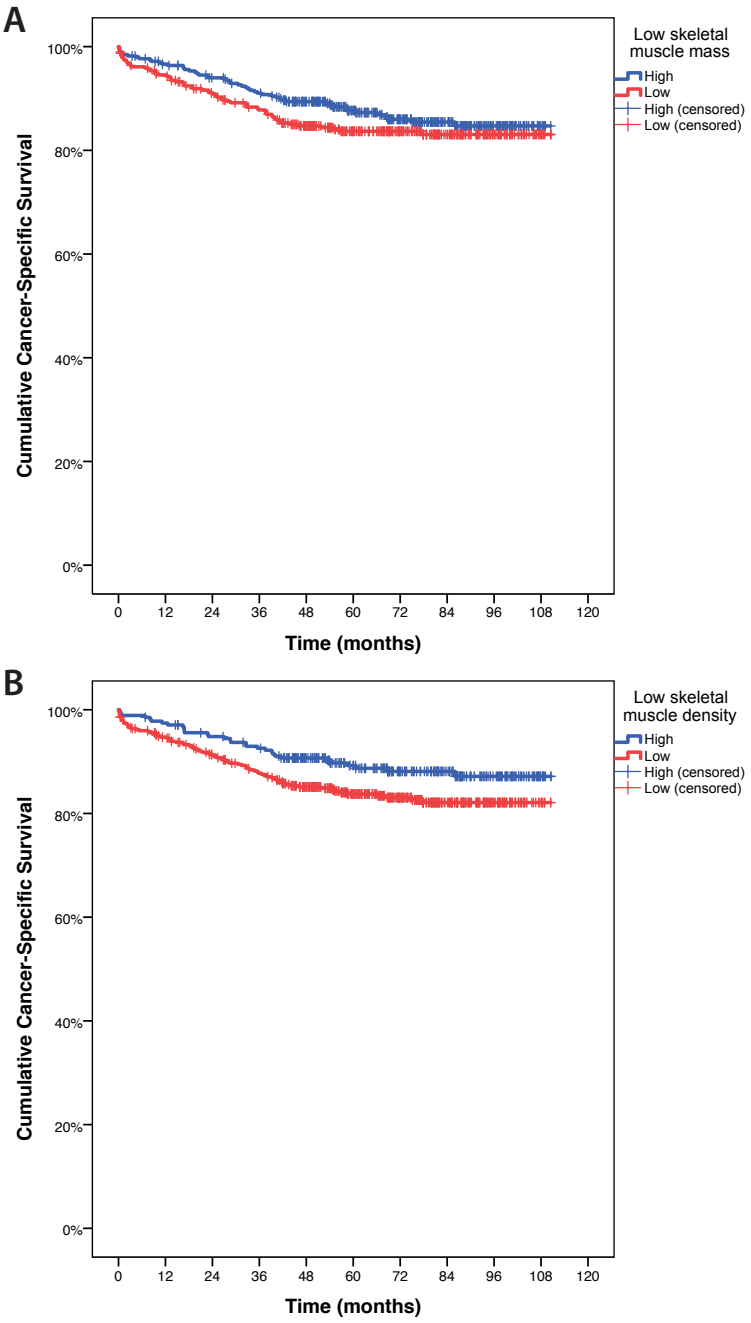

Figure 2. A. Kaplan-Meier curve for cancer-specific survival for patients with low skeletal muscle mass and high skeletal muscle mass (p=0.235). B. Kaplan-Meier curve for cancer-specific survival for patients with low skeletal muscle density and high skeletal muscle density (p=0.046).

Table 6. Cox proportional hazards analysis for overall survival, excluding those patients who died postoperatively (i.e. Comprehensive complication index 100) (n=797).

	HR (95% CI)	p-value	Adjusted HR (95% CI)	p-value
Skeletal muscle mass				
High	1	0.029	1	0.680
Low	1.35 (1.03-1.76)		1.06 (0.80-1.42)	
Low skeletal muscle density				
High	1	<0.001	1	0.600
Low	1.75 (1.29-2.36)		0.91 (0.65-1.29)	
Sex				
Female	1	0.399		
Male	0.89 (0.68-1.16)			
Age	1.07 (1.05-1.09)	<0.001	1.06 (1.05-1.08)	<0.001
BMI				
<20	1	0.186		
20-24.9	0.63 (0.32-1.25)	0.184		
≥25.0	0.63 (0.32-1.24)			
Charlson comorbidity index	1.26 (1.18-1.35)	<0.001	1.17 (1.09-1.27)	<0.001
ASA classification				
I	1	<0.001		
II	2.83 (1.66-4.82)	<0.001		
III	4.22 (2.37-7.51)	0.040		
IV	3.19 (1.06-9.62)	<0.001		
V	55.32 (7.15-427.98)			
CEA	1.00 (1.00-1.01)	0.308		
Type of surgery				
Open	1	0.006	1	0.678
Laparoscopic	0.67 (0.51-0.89)	0.811	1.07 (0.77-1.50)	0.017
Conversion	1.05 (0.69-1.62)		1.77 (1.11-2.82)	
Stoma				
No	1	<0.001	1	<0.001
Yes	1.64 (1.24-2.16)		2.02 (1.47-2.77)	
Blood loss (100 ml)	0.98 (0.95-1.02)	0.368		
Duration of surgery (hours)	0.86 (0.75-0.99)	0.033	0.83 (0.71-0.97)	0.021
Tumor stage				
I	1	0.135	1	0.036
II	1.31 (0.92-1.85)	0.001	1.48 (1.03-2.15)	<0.001
III	1.81 (1.29-2.55)		3.14 (2.11-4.68)	
Tumor location				
Colon	1	0.450		
Rectum	1.12 (0.84-1.50)			

	HR (95% CI)	p-value	Adjusted HR (95% CI)	p-value
Tumor grade				
Good	1	0.374	1	0.265
Moderate	0.86 (0.61-1.20)	0.010	0.82 (0.58-1.17)	0.051
Poor	1.79 (1.15-2.77)	0.685	1.59 (1.00-2.54)	0.747
Other	1.21 (0.48-3.04)		0.86 (0.34-2.18)	
Angioinvasive growth				
No	1	0.994		
Yes	1.00 (0.86-1.17)			
Neoadjuvant therapy				
No	1	0.824		
Yes	0.97 (0.71-1.32)			
Adjuvant therapy				
No	1	0.036	1	0.124
Yes	0.68 (0.47-0.97)		0.69 (0.43-1.11)	
Comprehensive complication index	1.02 (1.01-1.02)	<0.001	1.01 (1.00-1.02)	0.003

DISCUSSION

In this study, we found that low skeletal muscle mass and density were predictors for short-term outcome (i.e. postoperative complications, mortality, LOS, and discharge status) in patients undergoing curative intent resection of colorectal cancer, but not for long-term outcome (i.e. overall, cancer-specific, and disease-free survival).

Our results are in line with the study of Sabel *et al.*, which also found an association between psoas density and short-term, but not long-term outcome [17]. As in our study, an association was observed in univariate analysis, which did not hold after adjustment for age and Charlson Comorbidity Index, which are known to be two strong prognostic factors. A recent study also observed a univariable association between reduced skeletal muscle mass and overall survival, but not in multivariable analysis [26], whereas another study among 805 patients undergoing colorectal cancer surgery did find independent associations between myopenia (i.e. low skeletal muscle mass) and overall and disease-free survival [16]. However, the latter also included stage IV cancer patients. Another Japanese study among 220 patients also found an independent association between low skeletal muscle mass and overall and recurrence-free survival [15]. However, in these studies patients were classified as having myopenia using cut-off values which have been derived in a North-American cohort and the sex-specific lowest quartile for skeletal muscle mass, respectively. These cut-off values are not stratified for BMI [27]. We choose to use the more recently proposed cut-off values by Martin *et al.*, stratified for both sex and BMI [25]. Furthermore, we excluded stage IV patients, since their diagnosis and treatment greatly differ from patients with stage I-III (i.e. non metastasized) disease. Furthermore, the effect of proinflammatory cytokine levels and muscle wasting is probably more severe in stage IV patients compared with stage I-III patients.

The fact that low skeletal muscle mass and density are associated with short-term, but not long-term, outcome suggests an increased vulnerability of colorectal cancer patients towards stressors, such as surgery (i.e. frailty) [28]. Once patients have survived the postoperative period, the effects of low skeletal muscle mass and density disappear. Other risk factors, such as age and comorbidity, become more important for the clinical course of a given patient. We hypothesize that most cancer populations, such as stage I-III colorectal cancer patients, generally reach a functional status after treatment (i.e. surgery) that is close to pre-disease levels [29]. In contrast, other cancer populations undergoing cancer surgery including sicker patients, such as pancreatic or liver cancer patients, have an increased risk of spiraling down a vicious circle, which progressively

enhances physical impairment. Furthermore, the more dismal prognosis of these cancers may at least be one cause that could explain the prognostic value of low skeletal muscle mass and/or density in these patients [3].

The absence of a preoperatively elevated inflammatory response (i.e. elevated neutrophil to lymphocyte ratio), as well as laparoscopy, are associated with preservation of skeletal muscle mass after colorectal cancer surgery, whereas higher age, female gender, higher ASA classification are associated with a further decrease of skeletal muscle mass [30]. Particularly in the last three months of life in advanced colorectal cancer patients, a significant increase in high metabolic rate tissues (e.g., liver, tumor) and a concurrent increase in resting energy expenditure and decrease in peripheral tissue (i.e. skeletal muscle mass and adipose tissue) has been shown [31]. Future research should elucidate the value of skeletal muscle preservation or decrease over time to early predict survival or disease recurrence.

The association between low skeletal muscle mass and density, and complications after colorectal cancer surgery has been described before [2, 3, 5-8, 26, 32]. This association may explained in part by an increased inflammatory state in patients with low skeletal muscle mass and increased age and frailty [33-36]. As previously suggested [8], the preoperative period allows for prehabilitation of colorectal cancer patients that could reduce postoperative complications and mortality. Particularly in rectal cancer patients, the period of neoadjuvant chemo- and/or radiotherapy, may form a perfect time window, as such programs are proven to be safe in patients undergoing chemotherapy [37, 38]. Such an intervention should be multidimensional, including at least exercise therapy, nutritional support, and therapeutics (e.g., anti-inflammatory drugs or myostatin antagonists). Currently, multiple studies are being conducted investigating medication to reduce or reverse cancer-induced skeletal muscle depletion [39-41].

Counterintuitively, patients in the low skeletal muscle density group had a lower tumor stage compared with the high density group. However, a similar association was observed in a previous by Miyamoto *et al.* [14], whereas other studies did not find any significant differences in cancer stage within non-metastasized patients [7]. In our study population, these differences are explained by the significant differences in age. Furthermore, tumor burden in patients without distant metastases may be less important than other factors, such as tumor biology, as previously described [36]. A significantly lower proportion of patients with low skeletal muscle density or mass underwent adjuvant chemotherapy, which may be explained by the fact that these patients were considered less fit.

Although this is currently the largest, and the only multicenter, study investigating the effect of low skeletal muscle mass and density on postoperative outcome in colorectal cancer patients undergoing surgery, non-significant differences may still be the result of an underpowered study. Furthermore, some patients could not be included because of unavailability of eligible CT examinations or body height that was not recorded in patient charts. Especially in rectal cancer patients CT examinations were often lacking. This may have led to some selection bias. However since most important baseline characteristics (such as sex, age, comorbidity, and tumor stage) of excluded patients did not significantly differ from included patients, this bias is probably limited.

In conclusion, low skeletal muscle mass and density are associated with an impaired postoperative recovery and increased postoperative complication rate. No association was found with overall, cancer-specific, and disease-free survival. The preoperative period may be used to optimize patients preoperatively.

REFERENCES

1. van Vugt JL, Reisinger KW, Derikx JP, Boerma D, Stoot JH. Improving the outcomes in oncological colorectal surgery. World J Gastroenterol 2014; 20: 12445-57.
2. Margadant CC, Bruns ER, Sloothaak DA, van Duijvendijk P, van Raamt AF, van der Zaag HJ, et al. Lower muscle density is associated with major postoperative complications in older patients after surgery for colorectal cancer. Eur J Surg Oncol 2016; 42: 1654-9.
3. Levolger S, van Vugt JL, de Bruin RW, JN IJ. Systematic review of sarcopenia in patients operated on for gastrointestinal and hepatopancreatobiliary malignancies. Br J Surg 2015; 102: 1448-58.
4. Rolland Y, Abellan van Kan G, Gillette-Guyonnet S, Vellas B. Cachexia versus sarcopenia. Current opinion in clinical nutrition and metabolic care 2011; 14: 15-21.
5. Huang DD, Wang SL, Zhuang CL, Zheng BS, Lu JX, Chen FF, et al. Sarcopenia, as defined by low muscle mass, strength and physical performance, predicts complications after surgery for colorectal cancer. Colorectal Dis 2015; 17: O256-64.
6. Jones KI, Doleman B, Scott S, Lund JN, Williams JP. Simple psoas cross-sectional area measurement is a quick and easy method to assess sarcopenia and predicts major surgical complications. Colorectal Dis 2015; 17: O20-6.
7. Lieffers JR, Bathe OF, Fassbender K, Winget M, Baracos VE. Sarcopenia is associated with postoperative infection and delayed recovery from colorectal cancer resection surgery. Br J Cancer 2012; 107: 931-6.
8. Tegels JJW, Van Vugt JLA, Reisinger KW, Hulsewé KWE, Hoofwijk AGM, Derikx JPM, et al. Sarcopenia is highly prevalent in patients undergoing surgery for gastric cancer but not associated with worse outcomes. J Surg Oncol 2015; 112: 403-7.
9. Blauwhoff-Buskermolen S, Versteeg KS, de van der Schueren MA, den Braver NR, Berkhof J, Langius JA, et al. Loss of Muscle Mass During Chemotherapy Is Predictive for Poor Survival of Patients With Metastatic Colorectal Cancer. J Clin Oncol 2016; 34: 1339-44.
10. Jung HW, Kim JW, Kim JY, Kim SW, Yang HK, Lee JW, et al. Effect of muscle mass on toxicity and survival in patients with colon cancer undergoing adjuvant chemotherapy. Support Care Cancer 2015; 23: 687-94.
11. Barret M, Antoun S, Dalban C, Malka D, Mansourbakht T, Zaanan A, et al. Sarcopenia is linked to treatment toxicity in patients with metastatic colorectal cancer. Nutrition and cancer 2014; 66: 583-9.
12. Peng PD, van Vledder MG, Tsai S, de Jong MC, Makary M, Ng J, et al. Sarcopenia negatively impacts short-term outcomes in patients undergoing hepatic resection for colorectal liver metastasis. HPB (Oxford) 2011; 13: 439-46.

13. van Vledder MG, Levolger S, Ayez N, Verhoef C, Tran TC, IJzermans JN. Body composition and outcome in patients undergoing resection of colorectal liver metastases. Br J Surg 2012; 99: 550-7.
14. Miyamoto Y, Baba Y, Sakamoto Y, Ohuchi M, Tokunaga R, Kurashige J, et al. Negative Impact of Skeletal Muscle Loss after Systemic Chemotherapy in Patients with Unresectable Colorectal Cancer. PLoS One 2015; 10: e0129742.
15. Miyamoto Y, Baba Y, Sakamoto Y, Ohuchi M, Tokunaga R, Kurashige J, et al. Sarcopenia is a Negative Prognostic Factor After Curative Resection of Colorectal Cancer. Ann Surg Oncol 2015; 22: 2663-8.
16. Malietzis G, Currie AC, Athanasiou T, Johns N, Anyamene N, Glynne-Jones R, et al. Influence of body composition profile on outcomes following colorectal cancer surgery. Br J Surg 2016; 103: 572-80.
17. Sabel MS, Terjimanian M, Conlon AS, Griffith KA, Morris AM, Mulholland MW, et al. Analytic morphometric assessment of patients undergoing colectomy for colon cancer. J Surg Oncol 2013; 108: 169-75.
18. Buttner S, Lalmahomed ZS, Coebergh van den Braak RR, Hansen BE, Coene PP, Dekker JW, et al. Completeness of pathology reports in stage II colorectal cancer. Acta Chir Belg 2017; 117: 181-7.
19. Lalmahomed ZS, Coebergh van den Braak RR, Oomen MH, Arshad SP, Riegman PH, JN IJ, et al. Multicenter fresh frozen tissue sampling in colorectal cancer: does the quality meet the standards for state of the art biomarker research? Cell Tissue Bank 2017.
20. von Elm E, Altman DG, Egger M, Pocock SJ, Gotzsche PC, Vandenbroucke JP. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: guidelines for reporting observational studies. Lancet 2007; 370: 1453-7.
21. Charlson ME, Pompei P, Ales KL, MacKenzie CR. A new method of classifying prognostic comorbidity in longitudinal studies: development and validation. J Chronic Dis 1987; 40: 373-83.
22. Dindo D, Demartines N, Clavien PA. Classification of surgical complications: a new proposal with evaluation in a cohort of 6336 patients and results of a survey. Annals of surgery 2004; 240: 205-13.
23. Slankamenac K, Nederlof N, Pessaux P, de Jonge J, Wijnhoven BP, Breitenstein S, et al. The comprehensive complication index: a novel and more sensitive endpoint for assessing outcome and reducing sample size in randomized controlled trials. Annals of surgery 2014; 260: 757-62; discussion 62-3.
24. van Vugt JL, Levolger S, Gharbharan A, Koek M, Niessen WJ, Burger JW, et al. A comparative study of software programmes for cross-sectional skeletal muscle and adipose tissue measurements on abdominal computed tomography scans of rectal cancer patients. J Cachexia Sarcopenia Muscle 2017; 8: 285-97.

25. Martin L, Birdsell L, Macdonald N, Reiman T, Clandinin MT, McCargar LJ, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol 2013; 31: 1539-47.

26. Black D, Mackay C, Ramsay G, Hamoodi Z, Nanthakumaran S, Park KG, et al. Prognostic Value of Computed Tomography: Measured Parameters of Body Composition in Primary Operable Gastrointestinal Cancers. Ann Surg Oncol 2017.

27. Prado CM, Lieffers JR, McCargar LJ, Reiman T, Sawyer MB, Martin L, et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. Lancet Oncol 2008; 9: 629-35.

28. Makary MA, Segev DL, Pronovost PJ, Syin D, Bandeen-Roche K, Patel P, et al. Frailty as a predictor of surgical outcomes in older patients. Journal of the American College of Surgeons 2010; 210: 901-8.

29. Hamaker ME, Prins MC, Schiphorst AH, van Tuyl SA, Pronk A, van den Bos F. Long-term changes in physical capacity after colorectal cancer treatment. Journal of geriatric oncology 2015; 6: 153-64.

30. Malietzis G, Currie AC, Johns N, Fearon KC, Darzi A, Kennedy RH, et al. Skeletal Muscle Changes After Elective Colorectal Cancer Resection: A Longitudinal Study. Ann Surg Oncol 2016; 23: 2539-47.

31. Lieffers JR, Mourtzakis M, Hall KD, McCargar LJ, Prado CM, Baracos VE. A viscerally driven cachexia syndrome in patients with advanced colorectal cancer: contributions of organ and tumor mass to whole-body energy demands. The American journal of clinical nutrition 2009; 89: 1173-9.

32. Boer BC, de Graaff F, Brusse-Keizer M, Bouman DE, Slump CH, Slee-Valentijn M, et al. Skeletal muscle mass and quality as risk factors for postoperative outcome after open colon resection for cancer. Int J Colorectal Dis 2016; 31: 1117-24.

33. Malietzis G, Johns N, Al-Hassi HO, Knight SC, Kennedy RH, Fearon KC, et al. Low Muscularity and Myosteatosis Is Related to the Host Systemic Inflammatory Response in Patients Undergoing Surgery for Colorectal Cancer. Annals of surgery 2016; 263: 320-5.

34. Reisinger KW, Derikx JP, van Vugt JL, Von Meyenfeldt MF, Hulsewe KW, Olde Damink SW, et al. Sarcopenia is associated with an increased inflammatory response to surgery in colorectal cancer. Clin Nutr 2016; 35: 924-7.

35. Richards CH, Roxburgh CS, MacMillan MT, Isswiasi S, Robertson EG, Guthrie GK, et al. The relationships between body composition and the systemic inflammatory response in patients with primary operable colorectal cancer. PLoS One 2012; 7: e41883.

36. McSorley ST, Black DH, Horgan PG, McMillan DC. The relationship between tumour stage, systemic inflammation, body composition and survival in patients with colorectal cancer. Clin Nutr 2017.

37. Adamsen L, Quist M, Andersen C, Moller T, Herrstedt J, Kronborg D, et al. Effect of a multimodal high intensity exercise intervention in cancer patients undergoing chemotherapy: randomised controlled trial. Bmj 2009; 339: b3410.

38. van Waart H, Stuiver MM, van Harten WH, Geleijn E, Kieffer JM, Buffart LM, et al. Effect of Low-Intensity Physical Activity and Moderate- to High-Intensity Physical Exercise During Adjuvant Chemotherapy on Physical Fitness, Fatigue, and Chemotherapy Completion Rates: Results of the PACES Randomized Clinical Trial. J Clin Oncol 2015; 33: 1918-27.

39. Dingemans AM, de Vos-Geelen J, Langen R, Schols AM. Phase II drugs that are currently in development for the treatment of cachexia. Expert Opin Investig Drugs 2014; 23: 1655-69.

40. Currow D, Temel JS, Abernethy A, Milanowski J, Friend J, Fearon KC. ROMANA 3: A phase 3 safety extension study of anamorelin in advanced non-small cell lung cancer (NSCLC) patients with cachexia. Annals of oncology : official journal of the European Society for Medical Oncology / ESMO 2017.

41. Solheim TS, Laird BJA, Balstad TR, Stene GB, Bye A, Johns N, et al. A randomized phase II feasibility trial of a multimodal intervention for the management of cachexia in lung and pancreatic cancer. J Cachexia Sarcopenia Muscle 2017.

CHAPTER 9

Low Skeletal Muscle Density is Associated with Early Death in Patients with Perihilar Cholangiocarcinoma Regardless of Subsequent Treatment

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ABSTRACT

Background: Low skeletal muscle mass is associated with increased postoperative morbidity and worse survival following liver resection for perihilar cholangiocarcinoma (PHC). The objective of this study was to investigate skeletal muscle mass and density to predict survival of all patients with suspected PHC, regardless of treatment.

Methods: Baseline characteristics and parameters regarding disease and treatment were collected of all patients with PHC from 2002-2014. Skeletal muscle mass and density were measured at the level of the third lumbar vertebra on CT-scans. The association between skeletal muscle mass and density with overall survival (OS) was investigated using the Kaplan-Meier method en Cox survival.

Results: Median OS did not differ between patients with and without low skeletal muscle mass (p=0.203), whereas a significantly lower median survival was observed in patients with low (7.0 months, 95%CI 4.7-9.3) compared with patients with high (12.1, 95%CI 8.1-16.1) skeletal muscle density (p=0.004). Low skeletal muscle density was independently associated with decreased survival (HR 1.78, 95%CI 1.03-3.07, p=0.04) within the first six months, but not after six months (HR 0.68, 95%CI 0.44-1.07, p=0.093), after adjusting for age, tumor size, and suspected peritoneal or other distant metastases on imaging.

Conclusions: A time-dependent effect of skeletal muscle density on mortality was found in patients with PHC, regardless of subsequent treatment. Low skeletal muscle density may identify patients at risk for early death.

INTRODUCTION

The prognosis of patients with perihilar cholangiocarcinoma (PHC) is poor. After curative-intent resection the median survival is 19-39 months with a 5-year survival rate of 10-40% [1-3]. However, only about one in four patients with suspected PHC undergoes surgical resection. The majority of patients receives palliative treatment or best supportive care and have a median survival of only 6 months [4-6].

Multiple staging systems are available to predict prognosis in patients with (suspected) PHC [4, 7-9]. However, the majority of staging systems, such as the frequently used American Joint Committee on Cancer (AJCC) staging system, are applicable only to the minority of patients that undergo surgical resection [7]. Prognostic factors and models for all PHC patients regardless of treatment are rare [9].

Recently, low skeletal muscle mass (i.e. sarcopenia), as part of the cancer cachexia syndrome [10,11], has been introduced as a biomarker to predict treatment complications and worsened survival in gastrointestinal and hepato-pancreato-biliary cancer patients [12, 13]. It may detect malnutrition that is not visible otherwise [14]. Three studies found that preoperative low skeletal muscle mass was also associated with worse outcome in patients undergoing surgical resection of extrahepatic biliary cancer [15] and PHC [16, 17]. However, the association between sarcopenia and outcome in all PHC patients, regardless of treatment, and the prognostic value of skeletal muscle density remain unknown. Therefore, the aim of this study was to investigate the prognostic value of skeletal muscle mass and density at initial presentation in all patients with suspected PHC.

METHODS

Patients and data collection

All patients aged 18 years and older with suspected PHC who presented in Erasmus University Medical Center between 2002 and 2014 were identified, regardless of subsequent treatment. Demographics (e.g., age, sex) and clinical (e.g., presence of cholangitis or icterus at presentation), drainage, laboratory (e.g., bilirubin and carbohydrate antigen (CA) 19.9 levels), and treatment (e.g., chemotherapy, surgery) parameters were collected from digital medical records. Body mass index (BMI) was categorized according to the World Health Organization (WHO) classification [18]. PHC was defined as a mass at or near the biliary confluence, arising between the origin of the cystic duct and the segmental bile ducts [19]. In the absence of histopathological evidence, the diagnosis of suspected PHC was based on the opinion of the multidisciplinary hepatopancreatobiliary team based on clinical, radiological, endoscopic, and laboratory observations. Patients were excluded if benign disease was considered more likely during follow up. Patients who visited our center for drainage only once, or who already underwent treatment (i.e. surgery, chemotherapy) in the referral center, were also excluded.

Radiological examinations (i.e. contrast-enhanced computed tomography (CT) and/or magnetic resonance imaging (MRI) with or without cholangiopancreatography) were re-assessed by an experienced abdominal radiologist. Parameters assessed on imaging were tumor visibility, tumor size, Bismuth-Corlette classification [20], vascular involvement [8], lobar atrophy, lymph node status, and the presence of distant metastases. Based on these findings, the AJCC stage (7th edition) was assessed [19]. Stage I and II were analyzed together for the clinical AJCC stage, because T1 (stage I) and T2 (stage 2) cannot be distinguished on imaging alone. Vascular involvement was defined as tumor contact of at least 180 degrees around the portal vein and/or hepatic artery and its side branches. Involvement of lymph nodes along the cystic duct, common bile duct, hepatic artery and portal vein was classified as N1 and lymph node involvement around the aorta, caval vein, superior mesenteric artery, and celiac artery as N2 [19].

The municipal records database was checked for patients' survival status on May 9th 2016. A waiver was granted for this study from the Medical Ethical Committee of Erasmus University Medical Center.

Diagnostic work-up and treatment algorithm

The diagnostic work-up included, but was not limited to, imaging with contrast-enhanced CT, MRI, and/or MRI with cholangiopancreatography (MRCP). Typically, patients were only considered for exploratory laparotomy in the absence of metastatic disease and with involvement of less than 180 degrees of the hepatic artery. A resection was performed only when a complete resection (R0) was anticipated with an adequate functional liver remnant. Patients with metastatic or locally advanced (i.e. unresectable) disease were offered palliative chemotherapy. All other patients received best supportive care and palliative drainage.

Skeletal muscle mass and density

Skeletal muscle mass was measured on abdominal CT, using an in-house developed software package as previously described [21, 22]. In short, the cross-sectional skeletal muscle area (CSMA, in cm^2) was measured at the level of the third lumbar vertebra (L3) using a Hounsfield unit (HU) range of -30 to +150. The CSMA was adjusted for patients' height squared, as is conventional for body composition measurements, resulting in the skeletal muscle index (SMI, cm^2/m^2). The SMI is strongly correlated with total body skeletal muscle mass [23, 24]. Patients were classified as having low or normal/high SMI according to previously defined cut-off values in patients with PHC undergoing surgery [16]. The mean HU value of the CSMA, as a measure of skeletal muscle density that is closely related to muscle function [25, 26], was also recorded. Patients were divided in groups of low skeletal muscle density (below the median sex-specific value) and high skeletal muscle density (above the median sex-specific value) [27]. Sarcopenic obesity was defined as low skeletal muscle mass and BMI ≥30 kg^2/m^2 and myosteatotic obesity as low skeletal muscle density and BMI ≥30kg^2/m^2.

The first abdominal CT during the diagnostic work-up of PHC was used. Patients without available contrast-enhanced CT imaging during diagnostic work-up (i.e. before any treatment) were excluded, because treatment may influence skeletal muscle density measurements. Furthermore, patients of whom not all skeletal muscles at the level of L3 were depicted on CT were excluded from further analysis.

Statistical analyses

Continuous data are reported as median with interquartile range (IQR) or mean with standard deviation (SD), depending on the normality of the distribution. Normality of the distribution was tested using the Shapiro-Wilk test. Categorical variables are

reported as counts with percentages. Fischer's exact or chi-squared tests were used for the comparison of proportions, while continuous parameters were compared using Student's t-tests.

Overall survival (OS) was measured from the date of first presentation in the tertiary referral center. Survival estimates were compared using the Kaplan-Meier method and the log-rank test. Logistic regression analysis was used to compare the 3-months, 6-months, 1-year, 3-year, and 5-year survival rates. The association between skeletal muscle mass and density and survival was investigated using a multivariable Cox proportional hazard regression model, adjusting for known risk factors [9] and additional factors that were associated with impaired survival in univariable analysis. Hazard ratios (HRs) with 95% confidence interval (CI) were reported. Due to the large number of missing values, CA19-9 was not included in the final model. A subgroup analysis was performed in unresectable patients only. The effect of skeletal muscle density on the hazard was allowed to vary before and after 6 months of follow-up. Therefore, an interaction term between time and skeletal muscle density was included in the Cox regression model [28]. Two-tailed p-values below 0.05 were considered statistically significant. All statistical analyses were performed using SPSS for Windows (IBM Corp., Armonk, NY, USA), version 22.

RESULTS

Patient and tumor characteristics

In total, 285 patients who presented with suspected PHC in our center were identified. Of these 285 patients, a contrast-enhanced abdominal CT was available for 233 (81.8%) patients, who formed the study cohort. The median age was 66 (IQR 57-74) years, and 140 (60.1%) patients were male. The median BMI was 24.7 (IQR 22.4-26.8) kg^2/m^2 and 17 (7.3%) patients were obese. Body height was missing for 23 (9.9%) patients. Consequently, these patients were excluded from analyses requiring body height (i.e. skeletal muscle mass), but included in analyses using skeletal muscle density. Due to missing body height and/or weight, BMI was unknown for 50 (21.5%) patients. Twenty-eight (12.7%) patients had AJCC stage I/II, 50 (22.6%) stage III, and 143 (64.7%) stage IV (table 1). The median time between the first available contrast-enhanced CT performed for the suspicion on PHC and the first presentation in the tertiary referral center was 11 (IQR -3-25) days.

Treatment characteristics

Forty-one (17.6%) patients underwent surgical resection including two liver transplantation, and 72 (30.9%) patients underwent a laparotomy without resection. In these 72 patients, the intraoperative finding of metastases and locally advanced disease were the most common reasons for renouncing resection. The remaining 120 (51.5%) patients were considered unresectable at initial presentation, of whom 13 (11.3%) received palliative chemotherapy.

Low skeletal muscle mass

In total, 103 of the 210 (49.0%) of patients were considered to have low skeletal muscle mass (table 1). The median SMI was significantly higher in males (47.6 [IQR 43.6-51.9] cm^2/m^2) compared with females (38.3 [IQR 35.0-43.5] cm^2/m^2), $p<0.001$. Patients with low skeletal muscle mass were significantly older compared with patients with high skeletal muscle mass (69 versus 64 years, $p=0.040$) and had significantly higher C-reactive protein levels (19 versus 9, $p=0.002$) and CA19.9 levels (254 versus 162, $p=0.039$). Median BMI was significantly lower in patients with low versus high skeletal muscle mass (23.7 versus 25.7, $p<0.001$). Few patients had a BMI ≥30 kg^2/m^2 and low skeletal muscle mass (n=3) or low skeletal muscle density (n=11). Finally, the rate of metastatic disease at initial presentation was significantly higher in patients with low skeletal muscle mass (15.5% versus 4.7%, $p=0.009$) and non-significant differences were observed in treatment groups.

Table 1. Baseline and treatment characteristics of the total population and for patients with low and normal/high skeletal muscle mass and skeletal muscle density, respectively.

	All	Skeletal muscle mass			Skeletal muscle density		
	N=233	Low *N*=103	High *N*=107	p-value	Low *N*=131	High *N*=102	p-value
Patient characteristic							
Age, years (IQR)	66 (57-74)	69 (58-74)	64 (53-72)	0.040	72 (64-76)	59 (47-67)	<0.001
Sex							
Males (%)	140 (60.1)	56 (54.4)	71 (66.4)	0.076	81 (61.8)	43 (42.2)	0.537
Females (%)	93 (39.9)	47 (45.6)	36 (33.6)		50 (38.2)	59 (57.8)	
BMI, kg/m^2*	24.7 (22.5-26.8)	23.7 (21.3-25.7)	25.7 (23.9-27.9)	<0.001	25.2 (23.4-27.6)	24.4 (21.9-26.3)	0.032
ECOG (WHO) performance status$^+$							
1-2	215 (95.1)	94 (94.0)	99 (95.2)	0.706	118 (92.9)	97 (98.0)	0.079
3-4	11 (4.9)	6 (6.0)	5 (4.8)		9 (7.1)	2 (2.0)	
Weight loss (kg), yes	118 (52.4)	50 (50.5)	60 (57.7)	0.160	68 (53.5)	50 (51.0)	0.089
Jaundice at presentation, yes	182 (80.9)	85 (85.0)	79 (76.7)	0.133	105 (82.7)	77 (78.6)	0.437
Cholangitis at/before presentation or preoperative	129 (56.8)	8 (8.4)	5 (4.9)	0.320	69 (54.3)	60 (60.0)	0.392
CA19-9, kU/L$^\#$	220 (57-1297)	254 (129-1304)	162 (41-848)	0.039	232 (67-1351)	206 (44-877)	0.534
Albumin, g/L	38 (33-43)	38 (31-44)	39 (25-42)	0.750	37 (31-43)	38 (34-42)	0.669
Total bilirubin prior to drainage, µmol/L§	138 (61-225)	146 (77-230)	120 (53-199)	0.185	155 (86-234)	122 (57-208)	0.134
C-reactive protein, mg/L$^¥$	13 (7-29)	19 (9-37)	9 (5-20)	0.002	17 (9-30)	9 (5-21)	0.023
Thrombocytes, x10^9/L	284 (220-338)	287 (228-354)	281 (206-332)	0.266	259 (222-323)	307 (208-366)	0.174
Disease characteristic							
Suspected peritoneal or other distant organ metastases	26 (11.2)	16 (15.5)	5 (4.7)	0.009	18 (13.7)	8 (7.9)	0.164
Lymph node status on imaging†							
N0	122 (53.3)	54 (53.5)	60 (57.1)	0.858	70 (54.3)	52 (52.0)	0.267
N1	67 (29.3)	30 (29.7)	28 (26.7)		33 (25.6)	34 (34.0)	
N2	40 (17.5)	17 (16.8)	17 (16.2)		26 (20.2)	14 (14.0)	
Vascular involvement on imaging‡	148 (64.9)	63 (61.2)	67 (65.0)	0.564	86 (68.3)	62 (60.8)	0.240
Tumor size on imaging (millimetres)	22 (20-35)	25 (19-32)	27 (21-35)	0.386	26 (21-36)	26 (20-34)	0.292

	All	Skeletal muscle mass			Skeletal muscle density		
	N=233	Low *N*=103	High *N*=107	p-value	Low *N*=131	High *N*=102	p-value
Lobar atrophy on imaging	61 (26.5)	32 (31.1)	28 (26.7)	0.484	40 (31.2)	21 (20.6)	0.069
AJCC stage (radiological)							
I/II	28 (12.7)	12 (12.4)	14 (13.6)		14 (11.4)	14 (14.3)	
III	50 (22.6)	23 (23.7)	24 (23.3)	0.968	28 (22.8)	22 (22.4)	0.810
IV	143 (64.7)	62 (63.9)	65 (63.1)		81 (65.9)	62 (63.3)	
Blumgart classification							
Stage 1	60 (26.9)	28 (28.3)	27 (26.5)		34 (27.6)	26 (26.0)	
Stage 2	56 (25.1)	31 (31.3)	22 (21.6)	0.190	33 (26.8)	23 (23.0)	0.697
Stage 3	107 (48.0)	40 (40.4)	53 (52.0)		56 (45.5)	51 (51.0)	
Treatment							
Treatment groups							
Laparotomy with resection	41 (17.6)	18 (17.5)	23 (21.5)		17 (13.0)	24 (23.5)	
Laparotomy without resection	72 (30.9)	29 (28.2)	43 (40.2)	0.062	24 (18.3)	48 (47.1)	<0.001
No laparotomy, initially unresectable	120 (51.5)	56 (54.4)	41 (38.3)		90 (68.7)	30 (29.4)	
Chemotherapy	31 (14.3)	14 (14.1)	17 (17.5)	0.56	11 (8.8)	20 (21.7)	0.007

Categorical parameters are presented as counts (percentages) and continuous parameters as median (interquartile range). Abbreviations: BMI, Body Mass Index (* missing for 50 patients); ECOG, Eastern Cooperative Oncology Group ([+] missing for 6 patients); CA 19.9, carbohydrate antigen 19.9 ([#] missing for 77 patients); AJCC, American Joint Committee on Cancer.
[¥] missing for 92 patients
[§] Missing for 49 patients
[†] Involvement of lymph nodes was assessed according to the AJCC (7th edition).
[‡] Vascular involvement on imaging was defined as tumor contact of at least 180 degrees.

Low skeletal muscle density

Low skeletal muscle density was observed in 131 (56.2%) patients. Males had a significantly higher skeletal muscle density compared with females (38 [IQR 31-43] versus 31 [IQR 25-39] HU, p<0.001). While median BMI was higher in patients with high skeletal muscle mass, it was significantly higher in patients with low skeletal muscle density compared with high skeletal muscle density (25.2 versus 24.4, p=0.032). Furthermore, patients with low skeletal muscle density were older (70 versus 58 years, p<0.001), had a higher CRP level (17 versus 9, p=0.023), presented more with unresectable disease (87.0% versus 78.0%, p<0.001) and were less frequently treated with chemotherapy (8.8% versus 21.7%, p=0.007), table 1.

Survival

In total, 221 (94.8%) patients died during the study period. Median follow-up of the included patients who were alive at last follow-up was 25.3 (IQR 18.3-85.5) months. The 3-month, 6-month, 1-year, 3-year, and 5-year survival rates in the entire cohort were 79.0%, 60.9%, 42.1%, 7.7%, and 3.0%, respectively. Median OS for the entire cohort was 9.6 (4.1-20.5) months. Median OS for patients who underwent resection was 29.1 months compared with 7.9 months in patients who did not undergo resection (p<0.001).

Skeletal muscle mass and density and overall survival

The median OS did not differ between patients with low skeletal muscle mass and high skeletal muscle mass (10.8 months [95% CI 7.7-13.8] versus 10.3 months [95% CI 8.2-12.3], p=0.203), figure 1, whereas a significantly lower median survival was observed in patients with low compared with patients with high skeletal muscle density (7.0 months [95% CI 4.7-9.3] versus 12.1 months [95% CI 8.1-16.1], p=0.004), figure 2.

Lower survival rates were observed in patients with low skeletal muscle density compared with patients with high skeletal muscle density at 3 months (71.0% versus 89.2%, p=0.001; odds ratio [OR] 3.38, 95% CI 1.63-7.02, p=0.001), 6 months (51.9% versus 72.5%, p=0.003; OR 2.45 95% CI 1.11-4.26, p=0.002) and 1 year (35.1% versus 51.0%, p=0.015; OR 1.92, 95% CI 1.13-3.26, p=0.015), but not at 3 and 5 years (6.1% versus 9.8%, p=0.294, and 2.3% versus 3.9%, p=0.086, respectively). After adjusting for age, tumor size, and suspected peritoneal or other distant metastases on imaging, low skeletal muscle density was independently associated with decreased survival (HR 1.78, 95% CI 1.03-3.07, p=0.004) within the first six months, but not after six months (HR 0.68, 95% CI 0.44-1.07, p=0.093) (table 2). Similar results were observed when sex was added to the analyses and in a subgroup analysis in unresectable patients only (data not

shown). An incremental skeletal muscle density (entered as a continuous measure) was also independently associated with decreased survival within six months (HR 0.96, 95% CI 0.93-0.99, p=0.002), but not after six months.

Table 2. Cox proportional hazard regression analysis for factors associated with impaired survival.

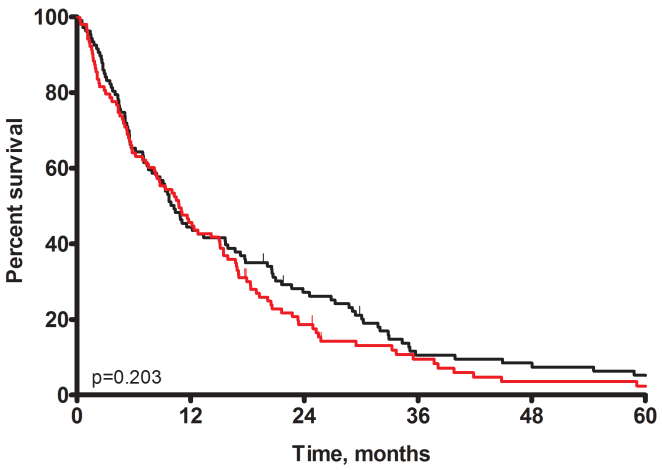
	Univariable		**Multivariable**	
	HR (95% CI)	**p-value**	**HR (95% CI)**	**p-value**
Age (years)	1.02 (1.01-1.03)	0.001	1.02 (1.01-1.04)	0.003
Sex				
Female	1 (ref)	0.945		
Male	1.01 (0.77-1.32)			
BMI ≥25 (kg/m^2)	1.04 (0.77-1.40)	0.803		
ECOG (WHO) performance status				
1-2	1 (ref)	0.403	1 (ref)	0.243
3-4	1.31 (0.69-2.48)		1.63 (0.72-3.69)	
Bilirubin >200 (µmol/L)	1.48 (1.00-2.19)	0.051	1.04 (0.67-1.60)	0.866
CA19.9 >1000 (kU/L)	1.87 (1.29-2.70)	0.001		
Albumin (g/dL)	0.99 (0.96-1.02)	0.429		
C-reactive protein (mg/L) ≥100	2.10 (1.05-4.18)	0.036		
Cholangitis before or at presentation	1.48 (0.84-2.60)	0.180		
Tumor size >3cm	2.31 (1.72-3.09)	<0.001	2.24 (1.60-3.15)	<0.001
Suspicious lymph nodes on imaging*				
N0	1 (ref)	0.046	1 (ref)	0.018
N1	1.37 (1.01-1.87)	0.033	1.57 (1.08-2.28)	0.134
N2	1.48 (1.03-2.13)		1.37 (0.91-2.06)	
Suspected distant metastases on imaging	1.46 (0.97-2.20)	0.072	3.74 (1.93-7.26)	<0.001
Lobar atrophy on imaging	1.04 (0.77-1.41)	0.793		
Vascular involvement on imaging[§]	1.44 (1.09-1.91)	0.011	1.30 (0.91-1.85)	0.150
Low skeletal muscle mass	1.99 (0.91-1.59)	0.204		
Low skeletal muscle density (<6 months)[#]	2.09 (1.34-3.27)	0.001	1.78 (1.03-3.07)	0.040
Low skeletal muscle density (≥6 months)[#]	1.20 (0.85-1.69)	0.306	0.68 (0.44-1.07)	0.093

Abbreviations: HR, Hazard Ratio; CI, Confidence Interval; BMI, Body Mass Index; ECOG, Eastern Cooperative Oncology Group; WHO, World Health Organization.

* Involvement of lymph nodes was assessed according to the AJCC (7th edition) [1].

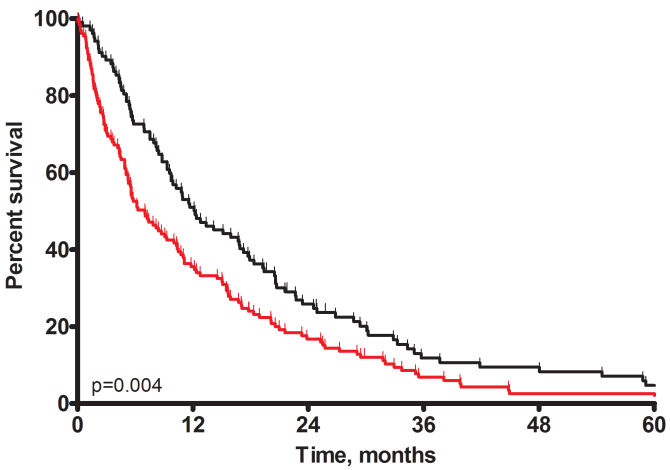
[#] The effect of skeletal muscle density on the hazard varied with time. Hence, an interaction term between skeletal muscle density and time was used to calculate the time-dependent effect of skeletal muscle density on the hazard.

[§] Vascular involvement on imaging was defined as tumor contact of at least 180 degrees around the portal vein and/or hepatic artery and its side branches.



High SMI	107	47	27	10	8	5
Low SMI	103	47	18	9	3	2

Figure 1. Kaplan-Meier survival curves for patients with high and low skeletal muscle mass (SMI).



High SMD	102	52	24	10	8	4
Low SMD	131	46	21	8	3	3

Figure 2. Kaplan-Meier survival curves for patients with high and low skeletal muscle density (SMD).

DISCUSSION

This study showed that low skeletal muscle density is an independent prognostic factor for overall survival within the first six months after initial presentation in patients with PHC. Skeletal muscle mass was not an independent prognostic factor.

This is the first study that investigated the association between skeletal muscle mass, skeletal muscle density and outcome in all patients with PHC in a unique Western series of patients with both resectable and unresectable PHC. In other tumors, such as follicular lymphoma, melanoma, and metastatic renal cell and gastric carcinoma no association between skeletal muscle mass and survival was shown, whereas skeletal muscle density was an independent prognostic factor [27, 29-32]. The similarity between these studies and the current study is the aggressive course of the disease, which may have led to the inability to accurately predict outcome.

An intriguing hypothesis described by Hayashi *et al.* is that a decrease in skeletal muscle density is detected earlier on CT than a decrease in skeletal muscle mass [31]. Recent studies show that skeletal muscle density is mainly correlated with intramuscular adipose tissue content [26], while low skeletal muscle mass results from limited muscle growth and increased muscle wasting [33]. The mechanisms leading to and effects of these two processes are probably different and further research on their pathophysiology is warranted. Tumor biology may play an important role, since the effects of skeletal muscle mass and density on outcome vary per tumor sort and within tumor sorts and altered body composition is associated with an elevated inflammatory response [34, 35].

The independent association between skeletal muscle mass and density has frequently been found in survival analyses of previous studies [12, 36]. Nevertheless, this is the first study to describe a time-dependent effect, independently of previously described risk factors for impaired survival in patients with PHC [9]. Although it should be assessed, time-dependency of covariates is often not assessed, leading to bias in survival analyses in a great part of literature [28]. Low skeletal muscle density influenced survival in the three to six months after initial diagnosis. However, this effect faded hereafter. This suggests that patients with the poorest survival are those with the lowest skeletal muscle density. Another reason why no effect was found after three and five years could have been the low median survival time (i.e. 7.9 months in unresectable and 29.1 months in resected patients), resulting in low patient numbers. Although we did not correct for treatment in multivariable analysis, we strongly believe that the model accurately reflects daily

practice. After all, the parameters assessed at first presentation greatly determine treatment and consequently (indirectly) correlate with survival. Our results should therefore be interpreted as valid in an "all-comers" patient population.

Notably, the rate of patients that underwent resection or received chemotherapy was lower in the low skeletal muscle density group. Furthermore, patients undergoing resection were significantly younger. These findings suggest a preoperative selection process of patients considered fit for surgery and chemotherapy. After all, none of the parameters representing tumor load (i.e. bilirubin level, CA19-9 level, vascular involvement, tumor size) that possibly may have influenced resectability, were significantly higher in patients with low skeletal muscle density. However, it should be noted that the median time interval between first presentation in the tertiary hospital and resection was 79 days. This time window may have led to further clinical deterioration and these findings should therefore be interpreted with caution. The significantly lower BMI and higher age in patients with low skeletal muscle density are in line with previous findings, as increasing age and adiposity are known for its association with intramuscular adipose tissue content [25, 37].

The majority of all prognostic models for PHC have been developed in patients undergoing surgical resection [9, 19]. However, the latter group forms the greatest number of patients with PHC, since only around a quarter of all patients undergo resection [4-6]. The value of skeletal muscle mass and density measurements to identify patients at risk for impaired outcome seems promising, particularly in hepaticopancreatobiliary cancer patients [12, 16, 17, 38]. Unfortunately, the number of patients who underwent surgical resection was too small to validate previously described findings regarding CT-assessed skeletal muscle mass and impaired outcome in patients with PHC undergoing surgery [16, 17]. Future studies should include low skeletal muscle density as a poor prognostic factor.

No international consensus has been reached on the definition of uniform cut-off points for CT-assessed skeletal muscle mass and density measurements yet [39], and various cut-off points are being used [16, 21, 23, 25]. Optimum stratification by means of the log-rank test is a method to identify optimal cut-off points for survival in a study cohort that has frequently been used [16, 21, 23, 25]. However, with a relatively small study cohort, it is highly questionable as to whether cut-off points can be reproduced in another sample of patients. Therefore, we choose to use the sex-specific median to group patients into low and high skeletal muscle density. Skeletal muscle density and survival was entered into the survival analysis as a continuous variable, since previous reports with large cohorts did not report sex differences in skeletal muscle attenuation [25]. Ideally, definitive cut-off points should be developed that are derived from healthy persons.

Although surgical resection is the main determinant of survival in perihilar cholangiocarcinoma, we opted to focus on the effects of pre-treatment parameters, in particular the impact of sarcopenia on patient survival. At the time of diagnosis, sarcopenia is easily measured using a spiral CT of the abdomen [21, 23-26]. Previous studies as well as the results of this study show that sarcopenia is heavily correlated with cancer stage and treatment, yet across all strata of treatment and cancer stage patients with sarcopenia perform worse [40, 41]. This indicates that, regardless of further choices sarcopenia is an independent predictor of outcome. By only taking into account the preoperative sarcopenia and radiological imaging, we therefore believe we have described the predictive ability of patient predisposition regardless of any treatment decisions. This predictive information could be valuable to improve informed clinical decision-making.

Some limitations of the current study should be acknowledged. A drawback is the retrospective character of the study design. Although a systematic search was performed in the electronic patient record, this may have led to selection bias. Furthermore, some variables had a high number of missing values. In 77 patients, for example, CA 19.9 was unknown, because this tumor marker assessment has not routinely been performed before 2010. Although only contrast-enhanced CTs were used for skeletal muscle mass and density measurements, possible differences as a consequence of the use of different CT scanners and scanning protocols in various hospitals, could not be precluded. The effect of contrast-enhancement on skeletal muscle mass and density measurements has not been evaluated. Skeletal muscle mass and density were measured at one time only (i.e. at initial presentation). Future studies could evaluate consecutive CT scans over time to allow the analysis of skeletal muscle mass and density changes over time and their association with treatment and mortality.

In conclusion, a time-dependent association between skeletal muscle density and mortality was found in patients with PHC, regardless of subsequent treatment. Low skeletal muscle density may identify patients with PHC at risk for early death. This finding should be validated in a larger, external cohort and future studies are needed to investigate the additional value of skeletal muscle density measurements in prognostic models.

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REFERENCES

1. Popescu I, Dumitrascu T. Curative-intent surgery for hilar cholangiocarcinoma: prognostic factors for clinical decision making. Langenbeck's archives of surgery / Deutsche Gesellschaft fur Chirurgie 2014; 399: 693-705.
2. Aljiffry M, Abdulelah A, Walsh M, Peltekian K, Alwayn I, Molinari M. Evidence-based approach to cholangiocarcinoma: a systematic review of the current literature. Journal of the American College of Surgeons 2009; 208: 134-47.
3. Groot Koerkamp B, Wiggers JK, Gonen M, Doussot A, Allen PJ, Besselink MG, et al. Survival after resection of perihilar cholangiocarcinoma-development and external validation of a prognostic nomogram. Annals of oncology : official journal of the European Society for Medical Oncology / ESMO 2016.
4. Jarnagin WR, Fong Y, DeMatteo RP, Gonen M, Burke EC, Bodniewicz BJ, et al. Staging, resectability, and outcome in 225 patients with hilar cholangiocarcinoma. Annals of surgery 2001; 234: 507-17; discussion 17-9.
5. Groot Koerkamp B, Wiggers JK, Allen PJ, Besselink MG, Blumgart LH, Busch OR, et al. Recurrence Rate and Pattern of Perihilar Cholangiocarcinoma after Curative Intent Resection. Journal of the American College of Surgeons 2015; 221: 1041-9.
6. Ruys AT, van Haelst S, Busch OR, Rauws EA, Gouma DJ, van Gulik TM. Long-term survival in hilar cholangiocarcinoma also possible in unresectable patients. World J Surg 2012; 36: 2179-86.
7. Edge SB, Compton CC. The American Joint Committee on Cancer: the 7th edition of the AJCC cancer staging manual and the future of TNM. Ann Surg Oncol 2010; 17: 1471-4.
8. Deoliveira ML, Schulick RD, Nimura Y, Rosen C, Gores G, Neuhaus P, et al. New staging system and a registry for perihilar cholangiocarcinoma. Hepatology 2011; 53: 1363-71.
9. Chaiteerakij R, Harmsen WS, Marrero CR, Aboelsoud MM, Ndzengue A, Kaiya J, et al. A new clinically based staging system for perihilar cholangiocarcinoma. Am J Gastroenterol 2014; 109: 1881-90.
10. Muscaritoli M, Bossola M, Aversa Z, Bellantone R, Rossi Fanelli F. Prevention and treatment of cancer cachexia: new insights into an old problem. Eur J Cancer 2006; 42: 31-41.
11. Fearon K, Strasser F, Anker SD, Bosaeus I, Bruera E, Fainsinger RL, et al. Definition and classification of cancer cachexia: an international consensus. Lancet Oncol 2011; 12: 489-95.
12. Levolger S, van Vugt JL, de Bruin RW, JN IJ. Systematic review of sarcopenia in patients operated on for gastrointestinal and hepatopancreatobiliary malignancies. Br J Surg 2015; 102: 1448-58.
13. van Vugt JL, Levolger S, de Bruin RW, van Rosmalen J, Metselaar HJ, JN IJ. Systematic Review and Meta-Analysis of the Impact of Computed Tomography-Assessed Skeletal Muscle Mass on Outcome in Patients Awaiting or Undergoing Liver Transplantation. Am J Transplant 2016; 16: 2277-92.

14. Barret M, Antoun S, Dalban C, Malka D, Mansourbakht T, Zaanan A, et al. Sarcopenia is linked to treatment toxicity in patients with metastatic colorectal cancer. Nutrition and cancer 2014; 66: 583-9.
15. Okumura S, Kaido T, Hamaguchi Y, Fujimoto Y, Kobayashi A, Iida T, et al. Impact of the preoperative quantity and quality of skeletal muscle on outcomes after resection of extrahepatic biliary malignancies. Surgery 2016; 159: 821-33.
16. Coelen RJ, Wiggers JK, Nio CY, Besselink MG, Busch OR, Gouma DJ, et al. Preoperative computed tomography assessment of skeletal muscle mass is valuable in predicting outcomes following hepatectomy for perihilar cholangiocarcinoma. HPB (Oxford) 2015; 17: 520-8.
17. Otsuji H, Yokoyama Y, Ebata T, Igami T, Sugawara G, Mizuno T, et al. Preoperative sarcopenia negatively impacts postoperative outcomes following major hepatectomy with extrahepatic bile duct resection. World J Surg 2015; 39: 1494-500.
18. Ogden CL, Carroll MD, Kit BK, Flegal KM. Prevalence of obesity and trends in body mass index among US children and adolescents, 1999-2010. Jama 2012; 307: 483-90.
19. Edge SB BD, Compton CC, Fritz AG, Greene FL, Trotti A, editors. AJCC cancer staging manual (7th ed). New York, NY: Springer; 2010.
20. Bismuth H, Corlette MB. Intrahepatic cholangioenteric anastomosis in carcinoma of the hilus of the liver. Surg Gynecol Obstet 1975; 140: 170-8.
21. van Vledder MG, Levolger S, Ayez N, Verhoef C, Tran TC, IJzermans JN. Body composition and outcome in patients undergoing resection of colorectal liver metastases. Br J Surg 2012; 99: 550-7.
22. van Vugt JL, Levolger S, Gharbharan A, Koek M, Niessen WJ, Burger JW, et al. A comparative study of software programs for cross-sectional skeletal muscle and adipose tissue measurements on abdominal computed tomography scans of rectal cancer patients. J Cachexia Sarcopenia Muscle 2016: [Epub ahead of print].
23. Prado CM, Lieffers JR, McCargar LJ, Reiman T, Sawyer MB, Martin L, et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. Lancet Oncol 2008; 9: 629-35.
24. Shen W, Punyanitya M, Wang Z, Gallagher D, St-Onge MP, Albu J, et al. Total body skeletal muscle and adipose tissue volumes: estimation from a single abdominal cross-sectional image. J Appl Physiol (1985) 2004; 97: 2333-8.
25. Martin L, Birdsell L, Macdonald N, Reiman T, Clandinin MT, McCargar LJ, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol 2013; 31: 1539-47.
26. Goodpaster BH, Kelley DE, Thaete FL, He J, Ross R. Skeletal muscle attenuation determined by computed tomography is associated with skeletal muscle lipid content. J Appl Physiol (1985) 2000; 89: 104-10.

27. Antoun S, Lanoy E, Iacovelli R, Albiges-Sauvin L, Loriot Y, Merad-Taoufik M, et al. Skeletal muscle density predicts prognosis in patients with metastatic renal cell carcinoma treated with targeted therapies. Cancer 2013; 119: 3377-84.

28. van Walraven C, Davis D, Forster AJ, Wells GA. Time-dependent bias was common in survival analyses published in leading clinical journals. J Clin Epidemiol 2004; 57: 672-82.

29. Chu MP, Lieffers J, Ghosh S, Belch AR, Chua NS, Fontaine A, et al. Skeletal muscle radio-density is an independent predictor of response and outcomes in follicular lymphoma treated with chemoimmunotherapy. PLoS One 2015; 10: e0127589.

30. Sabel MS, Lee J, Cai S, Englesbe MJ, Holcombe S, Wang S. Sarcopenia as a prognostic factor among patients with stage III melanoma. Ann Surg Oncol 2011; 18: 3579-85.

31. Hayashi N, Ando Y, Gyawali B, Shimokata T, Maeda O, Fukaya M, et al. Low skeletal muscle density is associated with poor survival in patients who receive chemotherapy for metastatic gastric cancer. Oncol Rep 2016; 35: 1727-31.

32. Van Rijssen LB, van Huijgevoort NC, Coelen RJ, Tol JA, Haverkort EB, Nio CY, et al. Skeletal Muscle Quality is Associated with Worse Survival After Pancreatoduodenectomy for Periampullary, Nonpancreatic Cancer. Ann Surg Oncol 2016.

33. Zhou X, Wang JL, Lu J, Song Y, Kwak KS, Jiao Q, et al. Reversal of cancer cachexia and muscle wasting by ActRIIB antagonism leads to prolonged survival. Cell 2010; 142: 531-43.

34. Reisinger KW, Derikx JP, van Vugt JL, Von Meyenfeldt MF, Hulsewe KW, Olde Damink SW, et al. Sarcopenia is associated with an increased inflammatory response to surgery in colorectal cancer. Clin Nutr 2016; 35: 924-7.

35. Richards CH, Roxburgh CS, MacMillan MT, Isswiasi S, Robertson EG, Guthrie GK, et al. The relationships between body composition and the systemic inflammatory response in patients with primary operable colorectal cancer. PLoS One 2012; 7: e41883.

36. Huang DD, Chen XX, Chen XY, Wang SL, Shen X, Chen XL, et al. Sarcopenia predicts 1-year mortality in elderly patients undergoing curative gastrectomy for gastric cancer: a prospective study. J Cancer Res Clin Oncol 2016; 142: 2347-56.

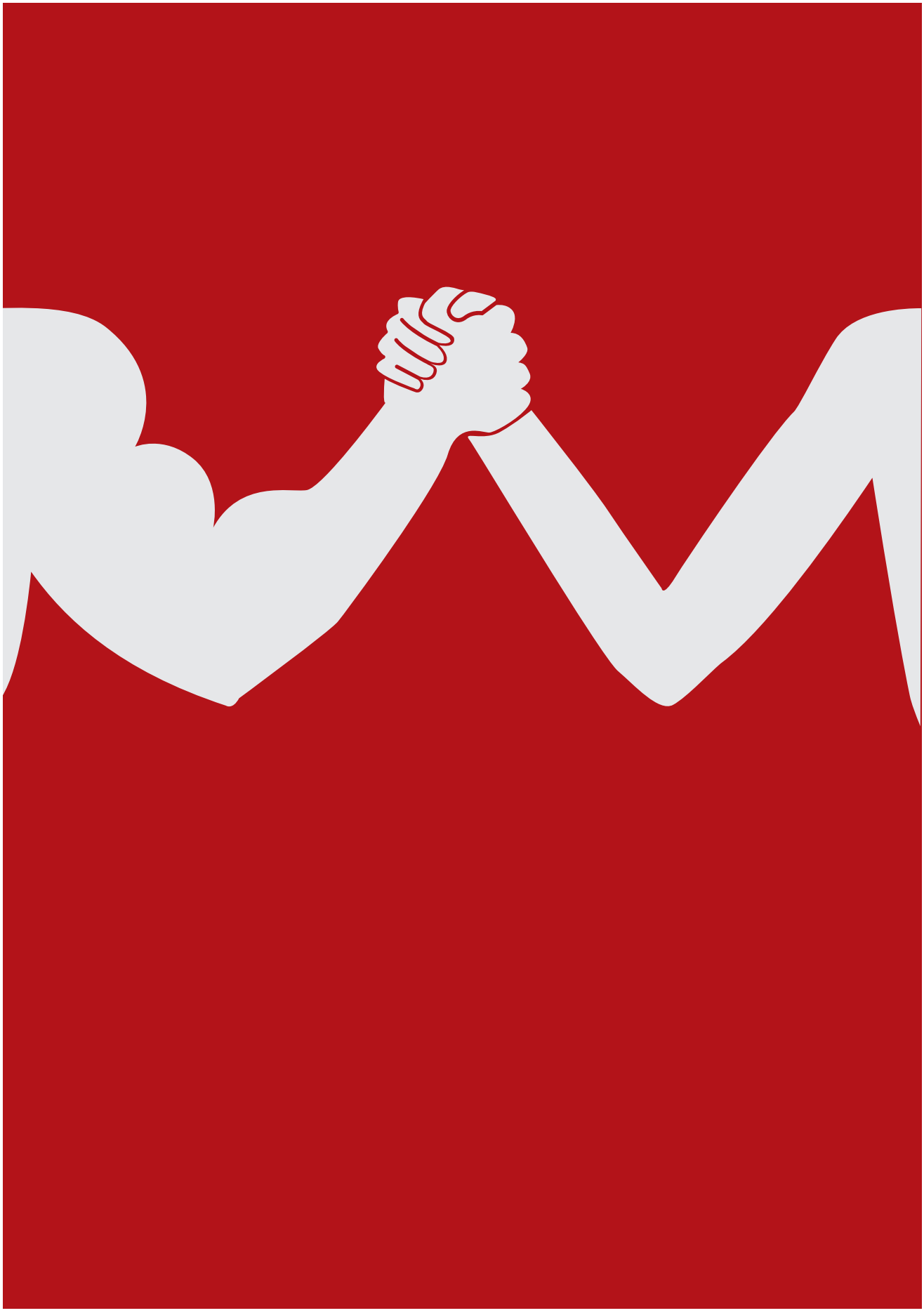
37. Anderson DE, D'Agostino JM, Bruno AG, Demissie S, Kiel DP, Bouxsein ML. Variations of CT-based trunk muscle attenuation by age, sex, and specific muscle. J Gerontol A Biol Sci Med Sci 2013; 68: 317-23.

38. Levolger S, van Vledder MG, Muslem R, Koek M, Niessen WJ, de Man RA, et al. Sarcopenia impairs survival in patients with potentially curable hepatocellular carcinoma. J Surg Oncol 2015; 112: 208-13.

39. Aubrey J, Esfandiari N, Baracos VE, Buteau FA, Frenette J, Putman CT, et al. Measurement of skeletal muscle radiation attenuation and basis of its biological variation. Acta Physiol (Oxf) 2014; 210: 489-97.

40. Kumar A, Moynagh MR, Multinu F, Cliby WA, McGree ME, Weaver AL, et al. Muscle composition measured by CT scan is a measurable predictor of overall survival in advanced ovarian cancer. Gynecol Oncol 2016; 142: 311-6.

41. Murton AJ, Maddocks M, Stephens FB, Marimuthu K, England R, Wilcock A. Consequences of Late-Stage Non-Small-Cell Lung Cancer Cachexia on Muscle Metabolic Processes. Clin Lung Cancer 2016.



PART IV

CONSEQUENCES OF LOW SKELETAL MUSCLE MASS IN LIVER TRANSPLANTATION

IV

CHAPTER 10

Systematic Review and Meta-Analysis of the Impact of Computed Tomography-Assessed Skeletal Muscle Mass on Outcome in Patients Awaiting or Undergoing Liver Transplantation

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ABSTRACT

Liver transplant outcome has improved considerably as a direct result of optimized surgical and anesthesiological techniques and organ allocation programs. As there is still a shortage of human organs, strict selection of transplant candidates remains of paramount importance. Recently, CT-assessed low skeletal muscle mass (i.e. sarcopenia) was identified as a novel prognostic parameter to predict outcome in liver transplant candidates. A systematic review and meta-analysis on the impact of CT-assessed skeletal muscle mass on outcome in liver transplant candidates were performed according to the PRISMA-guidelines. Nineteen studies, including 3803 patients in partly overlapping cohorts, fulfilled the inclusion criteria. The prevalence of sarcopenia ranged from 22.2-70%. An independent association between low muscle mass and post-transplantation and waiting list mortality was described in four of the six and six of the eleven studies, respectively. The pooled hazard ratios of sarcopenia were 1.84 (95% CI 1.11-3.05, p=0.02) and 1.75 (95% CI 0.99-3.00, p=0.05), for post-transplantation and waiting list mortality, respectively, independent of Model for End-stage Liver Disease (MELD) score. Less consistent evidence suggested a higher complication rate, particularly infections, in sarcopenic patients. In conclusion, sarcopenia is an independent predictor for outcome in liver transplantation patients and could be used for risk assessment.

INTRODUCTION

As human organ shortage remains prevalent, strict selection of transplant candidates is of paramount importance. The combination of waiting list mortality and post-transplantation survival are key deciding factors in waiting list placement. Currently, the Model for End-stage Liver Disease (MELD) score, a validated risk-based system that predicts waiting list mortality, is used to allocate donor livers [2]. Although the introduction of the MELD-score has led to a decreased number of patients on the waiting list, shortened waiting time and decreased waiting list mortality despite increasing disease severity [3], objective parameters reflecting a patient's nutritional and functional status in particular are lacking and attempts have been made to modify and improve the MELD-score [4, 5]. Frailty, the inability to adequately respond to stressors (i.e. surgery), for instance, has been identified as a prevalent syndrome in liver transplant candidates that strongly predicts waiting list mortality [6].

Skeletal muscle wasting (i.e. sarcopenia), which is a common syndrome in chronic diseases such as liver failure, is a key feature of frailty. The association between sarcopenia and treatment outcomes, such as complications and survival, using single-slice computed tomography (CT) based measurements has recently been described in various patient groups [7]. Sarcopenia is frequently found to be an independent predictor for treatment outcome, and is considered to be a stronger predictive marker than conventional risk factors, such as age and American Society of Anesthesiologists (ASA) classification [8, 9]. However, study results remain inconclusive. Therefore, the aim of this study was to systematically review the impact of CT-based skeletal muscle measurements on outcome in patients awaiting or undergoing liver transplantation.

METHODS

The study was registered in the PROSPERO International prospective register of systematic reviews (CRD42015019086) [8]. *A priori* defined eligibility criteria were established. All original studies that investigated the influence of skeletal muscle mass by means of abdominal CT in patients who underwent liver transplantation or were registered on the waiting list were identified by a systematic search performed in EMBASE, PubMed, and Web of Science, which was limited to English papers published between January 2000 and February 2015. The following search terms were used: ('sarcopenia':de,ab,ti OR 'analytic morphomics':de,ab,ti OR 'body composition':de,ab,ti OR 'muscle depletion':de,ab,ti OR 'muscle mass':de,ab,ti OR 'psoas area':de,ab,ti OR 'myopenia':de,ab,ti OR 'core muscle':de,ab,ti OR 'lean body mass':de,ab,ti OR 'muscular atrophy':de,ab,ti) AND ('liver transplantation':de,ab,ti). Similar queries were used for PubMed and Web of Science. The systematic review was performed according to the Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) guidelines [10].

Eligibility of studies and assessment of methodological quality

Duplicate records were removed and all abstracts were independently screened by two investigators to determine eligibility for further analysis. All abstracts describing the prevalence or predictive value for complications and survival of sarcopenia in patients awaiting or undergoing liver transplantation were further assessed. Studies that measured muscle mass with other means than CT were excluded. Only original studies were included. Case reports, review articles, opinion articles and experimental studies were excluded. The remaining full-text articles were subsequently retrieved and independently screened by two investigators. All articles within the inclusion criteria were included in the systematic review. The included full-text articles were screened for additional relevant references. The methodological quality of the included studies was independently assessed by two investigators using the Newcastle-Ottawa Quality Assessment Scale for Cohort Studies for each *a priori* defined outcome measure [11]. This is a ten-point scale, with 0 being poorest quality and 9 being highest quality. Quality assessment was performed separately for short and long term outcomes.

Data extraction

Two investigators independently extracted data regarding study design and results, including: age, gender distribution, patient selection, indication for liver transplantation or disease etiology, Body Mass Index (BMI), albumin level, MELD-score, presence of cirrhosis, details on skeletal muscle mass measurement methods, prevalence of

sarcopenia, waiting list mortality, post-transplantation mortality and complications, length of intensive care unit (ICU) and hospital stay, graft survival, and overall survival. Relevant information for the meta-analyses that could not be extracted from the articles was requested from the corresponding authors and when provided, included in the review. If not stated otherwise, results from multivariable analyses were used for the interpretation of the data.

Statistical analysis

All outcomes are reported as in the original articles. A meta-analysis was performed using Review Manager 5.3 (The Nordic Cochrane Center, Copenhagen, Denmark). Data are presented as hazard ratios (HR) with 95% confidence intervals (CI). If not stated otherwise, results of adjusted analyses were used. Random effects models were used to calculate summary estimates and to adjust for potential heterogeneity. Studies were weighted according to the inverse of the variance of the log hazard ratio. Overall effects were assessed using the Z-test and heterogeneity was tested using Cochran's chi-square test. The I^2 statistic was used to assess heterogeneity, which was defined as low, moderate, or high with I^2 values above 25%, 50%, and 75%, respectively [12]. If a research group contributed multiple studies with (partly) overlapping cohorts or relevant data was missing in the articles, the research group was contacted to provide additional data. If this data could not be provided, only the most relevant study was entered into the meta-analysis. Two-sided p-values <0.05 were considered statistically significant.

RESULTS

Of the 470 records that were found on February 3rd, 2015, 28 full text articles were considered potentially relevant (figure 1). From these 28 records, eight studies assessed muscle mass with means other than CT and one study was performed in another population than patients awaiting or undergoing liver transplantation. The remaining nineteen studies, including 3803 patients, were included in this systematic review [13-30]. Cross-referencing yielded no additional records.

table 1 shows the population characteristics and the quality of the enrolled studies. The main indications for liver transplantation were viral liver infections (i.e. hepatitis B and C), followed by alcoholic liver cirrhosis. Around 65% was male and the mean age was 52 to 62 years. The median MELD-score ranged from 9-21, the median albumin level from 2.8 to 3.4 g/dl, and median BMI from 24.0 to 29.4 kg/m^2. Eight studies included cirrhosis patients only [13, 16, 18, 23-25, 27, 28], of which one study Child Pugh A patients only [28].

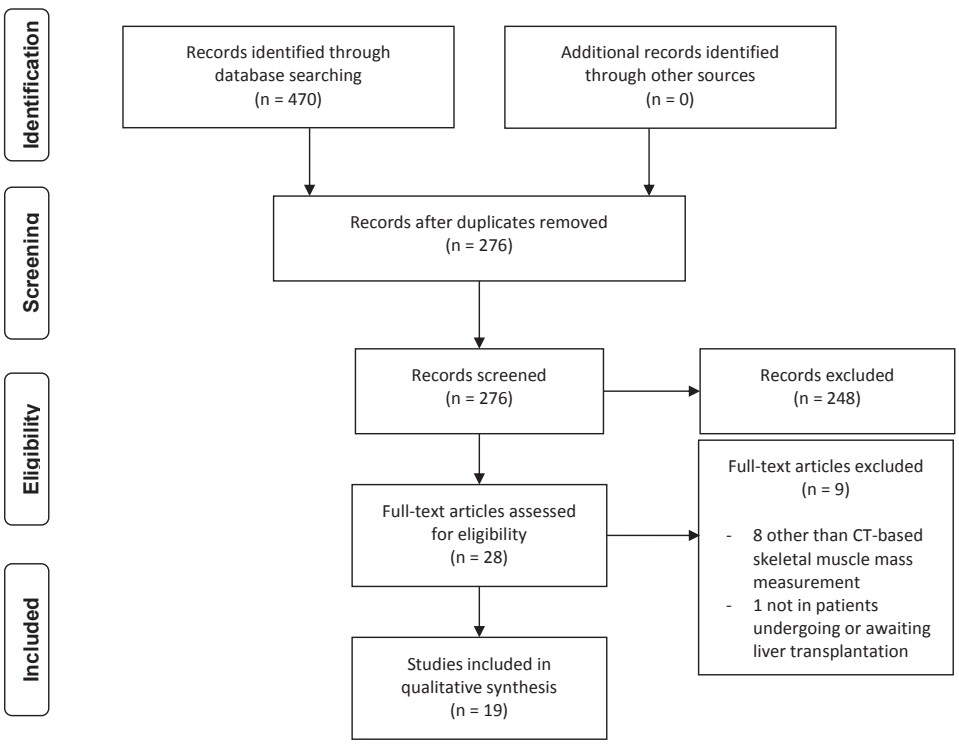


Figure 1. PRISMA Flow Chart of included studies.

Table 1. Study population characteristics.

Author, Year	Patient selection	*n* (%♀)	Age	BMI (kg/m²)	MELD-score	Albumin (g/dl)	LT indication	Quality points*	
								C	S
Bergerson, 2014 [1]	All patients undergoing LT because of alcohol, NASH or PSC cirrhosis (2000 – 2012)	40 (35)	57†	29†	15†	3.0†	NASH 53% PSC 25% Alcoholic 23% HCC 35%	n/a	n/a
Cruz, 2013 [2]	Adults evaluated for LT (Jan 2005 – Dec 2008)	234 (33)	55†	28†	21†	3.0†	HBV/HCV 25% Alcoholic 24% Alcoholic + HBV/HCV 12% NASH 12% Autoimmune/PSC/PBC 11% Fulminant liver failure 5% Other 11% HCC n/a	n/a	6/9
DiMartini, 2013 [3]	First time LT without transplantation of other organs (Jan 2005 – Dec 2008)	338 (34)	55†	28†	20†	3.0†	HBV/HCV 27% Alcoholic 23% NASH 14% Autoimmune/PSC/PBC 12% Alcohol + HBV/HCV 9% Fulminant liver failure 4% Other 11% HCC n/a	7/9	7/9
Durand, 2014 [4]	All consecutive patients with cirrhosis listed for deceased donor LT (2002 – 2011)	562 (19)	53†	26†	16†	N/a	Alcoholic 42% HCV 30% HBV 15% Biliary disease 5% Other 8% HCC 46%	n/a	3/9
Englesbe, 2010 [5]	Adult patients undergoing LT (2002 – 2008)	163 (37)	52†	28†	19†	2.8†	HCV 35% Alcoholic 12% PSC 10% PBC 6% Other 25% HCC 13%	n/a	7/9
Giusto, 2015 [6]	Adult patients with liver cirrhosis under evaluation for LT without acute liver failure and HCC beyond Milan criteria (2011 – 2013)	59 (22)	59‡	25‡	HCC: 11‡ No HCC: 16‡	N/a	HBV/HCV 56% Alcoholic 22% Other 22% HCC 41%	n/a	7/9
Hamaguchi, 2014 [7]	Adult patients undergoing LDLT (Jan 2008 – Oct 2013)	200 (53)	54‡	N/a	18‡	N/a	HBV/HCV 19% PSC/PBC 17% Other 31% HCC 34%	n/a	5/9

Author, Year	Patient selection	*n* (%♀)	Age	BMI (kg/m²)	MELD-score	Albumin (g/dl)	LT indication	Quality points*	
								C	S
Krell, 2013 [8]	Adult patients undergoing LT (June 2002 – Aug 2008)	207 (38)	52†	T1#: 27† T2#: 28† T3#: 29†	T1#: 23† T2#: 19† T3#: 18†	N/a	HBV/HCV 30% Autoimmune/PBC/PSC 23% Alcoholic 15% NASH 4% Fulminant liver failure 2% Other 12% HCC 25%	5/9	n/a
Lee, 2014 [9]	Adult patients undergoing LT (2000 – 2011)	325 (39)	52†	T1#: 27† T2#: 28† T3#: 30†	18†	T1#: 2.9† T2#: 2.8† T3#: 2.9†	Cirrhosis 40% HCV 29% HCC 39%	6/9	6/9
Masuda, 2014 [10]	Patients undergoing LDLT (Nov 2003 – Dec 2011)	204 (50)	54†	24†	≥20: S: 24% NS: 10%	N/a	HBV/HCV 63% PBC 13% Alcoholic 5% Other 19% HCC n/a	6/9	5/9
Meza-Junco, 2013 [11]	Consecutive patients with HCC and cirrhosis evaluated for LT	116 (16)	58†	29†	9†	3.4†	HBV/HCV 60% Alcoholic + HCV 20% Alcoholic 11% NASH 7% Other 3% HCC 100%	n/a	6/9
Montano-Loza, 2012 [12]	Consecutive patients with cirrhosis evaluated for LT (10% underwent LT)	112 (30)	54†	28†	13†	3.1†	HBVHCV 30% Alcoholic 22% Autoimmune/PBC/PSC 19% Alcoholic and HCV 16% Other 13% HCC n/a	n/a	7/9
Montano-Loza, 2014 [13]	Cirrhosis patients undergoing LT (2000 – 2012)	248 (32)	55†	S: 25† NS: 29†	20†	S: 3.3† NS: 3.4†	HBV/HCV 60% Alcoholic 19% Autoimmune/PBC/PSC 15% NASH 6% Other 1% HCC 39%	5/9	7/9
Tandon, 2012 [12]	Adult patients on the LT waiting list without HCC, acute liver failure, prior LT, multivisceral LT, LRLT (Feb 2005 – Nov 2009)	142 (40)	53‡	27‡	15‡	3.0‡	HCV + alcoholic 38% Autoimmune/PBC/PSC 25% Alcoholic 20% Cryptogenic/NAFLD 11% Other 7% HCC 0%	n/a	6/9
Toshima, 2015 [14]	LDLT recipients (Nov 2003 – Dec 2011)	143 (48)	S: 55† NS: 55†	24†	S: 17† NS: 13†	N/a	N/a	6/9	n/a

Author, Year	Patient selection	*n* (%♀)	Age	BMI (kg/m²)	MELD-score	Albumin (g/dl)	LT indication	Quality points*	
								C	S
Tsien, 2014 [15]	Adult cirrhosis patients undergoing LT (Jul 2009 – Jul 2011)	53 (23)	57†	29†	13†	3.3†	Viral 42% Alcoholic + viral 23% NASH 8% Other 28% HCC 64%	4/9	4/9
Valero, 2015 [16]	Child Pugh A patients undergoing hepatic resection or OLT for HCC or ICC (2000 – 2013)	96 (39)	62†	27†	10†	3.7†	29% underwent LT (100% HCC) ICC 30% HCC 70%	6/9	6/9
Waits, 2014 [17]	Adult patients who received liver transplants from deceased donors (2000 – 2011)	348 (38)	51†	T1¶: 27† T2¶: 28† T3¶: 29†	T1¶: 20† T2¶: 17† T3¶: 18†	T1¶: 2.9† T2¶: 2.8† T3¶: 2.8†	HCV 38% HCC 27%	n/a	6/9
Yadav, 2015 [18]	All patients listed for LT (Jul 2008 – Jul 2011)	213 (39)	55†	29† S: 24† NS: 29†	16†	3.3†	HCV 44% Alcoholic 16% NASH 14% PBC/PSC 8% Cryptogenic 6% Other 12% HCC n/a	n/a	7/9

† mean. ‡ median. * Scored with the Newcastle-Ottawa quality assessment scale for cohort studies, on a scale of 0 to 9, with 0 being poorest quality and 9 being highest quality. Quality assessment was performed separately for short and long term outcomes. # Tertiles based on skeletal muscle mass. ¶ Tertiles (young, middle, oldest) based on chronological age (psoas area, psoas density and abdominal aneurysmal calcifications). Abbreviations: LT; Liver Transplantation, C; complications, S; survival, LDLT; Living Donor Liver Transplantation, OLT; Orthotropic Liver Transplantation, HCC; Hepatocellular Carcinoma (either primary etiology or concomitant); BMI; Body Mass Index (kg²/m²), MELD; Model For End-Stage Liver Disease Score, S; patients with sarcopenia, NS; patients without sarcopenia, N/a; Not available. NASH; Nonalcoholic Steatohepatitis, PSC; Primary Sclerosing Cholangitis, HBV; Hepatitis B Virus, HCV; Hepatitis C Virus, PBC; Primary Biliary Cirrhosis, LRLT; living related liver transplantation.

Definitions and prevalence of sarcopenia

A great variety in skeletal muscle measurement methods and definitions used to classify patients as sarcopenic or non-sarcopenic was observed. The methods of muscle measurement and sarcopenia definitions that were used are summarized in table 2. Nine studies reported the cross-sectional muscle area with corresponding skeletal muscle index [13-15, 18, 23-25, 30], whereas the psoas area was reported in eight studies [16, 17, 19, 20, 22, 26-28] and the dorsal muscle group area in one study [21]. One study calculated the morphometric age (calculated with total psoas area, psoas density and abdominal aortic calcifications) [29]. The mean skeletal muscle index ranged from 43.0 cm^2/m^2 to 54.3 cm^2/m^2 [14, 30]. The prevalence of sarcopenia was reported in seventeen studies [13-15, 17-28, 30] and ranged from 22.2% [30] to nearly 70% [14]. The prevalence greatly depended on the definition used. All studies that reported the prevalence of sarcopenia separately for males and females, reported a higher prevalence among males [13, 15, 18, 22-26, 30].

Waiting list mortality

Four [16, 23, 24] of the six [16, 18, 23, 24, 30] studies investigating the association between skeletal muscle mass and mortality among patients being evaluated for or awaiting liver transplantation found an independent association. All details about survival rates and times can be found in table 3. The forest plot in figure 2a shows the meta-analysis of the association between sarcopenia and waiting list mortality with a pooled hazard ratio (HR) of 1.75 (95% CI 0.99-3.00, p=0.05) and low heterogeneity between studies (I^2=33%). Nevertheless, the evidence is limited, because three of the four studies with positive outcome were performed in one center [23, 24].

In the study of Durand *et al.*, an increasing transversal psoas muscle thickness corrected for height was associated with reduced mortality in both a pre-MELD cohort (HR 0.92 [95% CI 0.86-0.98], p=0.02) and MELD-era cohort (HR 0.86 [95% CI 0.78-0.94], p=0.001). Furthermore, the discrimination for waiting list mortality of the MELD-psoas area score was superior over the MELD-score and MELDNa-score (i.e. MELD-score with the addition of serum sodium), particularly in patients with a MELD-score ≤25 or refractory ascites [16]. Waiting list mortality was also greater among sarcopenic patients compared with non-sarcopenic patients in the study of Tandon *et al.* (log-rank p=0.04), and sarcopenia was an independent predictor of overall mortality in multivariable analysis (HR 2.36 [95% CI 1.23-4.53], p=0.009) [24]. Remarkably, outcome in sarcopenic patients with a low MELD-score (<15) was similar as for patients with a high MELD-score (≥ 15) with or without sarcopenia. In subgroup analyses, sarcopenia remained associated with mortality in patients with a low MELD-score (log rank p=0.02), whereas it was not in patients with a high MELD-score (log rank p=0.59). None of the other included studies performed comparable subgroup analyses.

Table 2. Methods used to measure skeletal muscle mass, definitions used to classify patients as sarcopenic and the prevalence of sarcopenia within studies.

Author, Year	Muscles measured	Software	Level	Cut off values / definition	Muscle area / density	Sarcopenia prevalence
Bergerson, 2014	CSA (SMI)	MITK software package	L3	♀ 38.5 cm^2/m^2 ♂ 52.4 cm^2/m^2	♀ 41.9 $cm^2/m^{2\dagger}$ ♂ 52.2 $cm^2/m^{2\dagger}$	♀ 43% ♂ 62%
Cruz, 2013	CSA (SMI)	SliceOmatic	Closest to L3-L4 disc space	♀ 38.5 cm^2/m^2 ♂ 52.4 cm^2/m^2	43.0 $cm^2/m^{2\dagger}$	Nearly 70% ♂ 76%
DiMartini, 2013	CSA (SMI)	SliceOmatic	Closest to L3-L4 disc space	♀ 38.5 cm^2/m^2 ♂ 52.4 cm^2/m^2	43.8 $cm^2/m^{2\dagger}$ ♀ 38.5 $cm^2/m^{2\dagger}$ ♂ 46.5 $cm^2/m^{2\dagger}$	68%
Durand, 2014	APMT, TPMT (right psoas muscle)	N/a	N/a	N/a (continuous parameter used)	N/a	N/a
Englesbe, 2010	TPA, PD	MATLAB	L4	Sex specific tertiles	TPA: 19.6 $cm^{2\dagger}$ PD: 101.0HU†	33% (lowest tertile)
Giusto, 2015	CSA (SMI)	Leonardo Syngo	Closest to L3-L4 disc space	♀ 38.5 cm^2/m^2 ♂ 52.4 cm^2/m^2	♀ 36.0 $cm^2/m^{2\ddagger}$ ♂ 49.9 $cm^2/m^{2\ddagger}$	76% ♀ 69% ♂ 78%
Hamaguchi, 2014	TPA (PMI), IMAC	Aquarius NET server	At level of subfascial muscular tissue in multifidus muscle	PMI: ♀ 4.1 cm^2/m^2 ♂ 6.7 cm^2/m^2 IMAC: ♀ -0.2 HU ♂ -0.4 HU	N/a	Low TPI: 44% High IMAC: 45%
Krell, 2013	TPA	MATLAB	L4	Sex specific tertiles	High TPA: ♀ 19.8 $cm^{2\dagger}$ ♂ 29.2 $cm^{2\dagger}$ Low TPA: ♀ 9.5 $cm^{2\dagger}$ ♂ 19.8 $cm^{2\dagger}$	33% (lowest tertile)
Lee, 2014	DMG*, PA	MATLAB	T12, L4	Sex specific tertiles	DMG: ♀ 25.3 $cm^{2\dagger}$ ♂ 33.9 $cm^{2\dagger}$ TPA: Low: 12.8 $cm^{2\dagger}$ High: 27.9 $cm^{2\dagger}$	33% (lowest tertile)
Masuda, 2014	TPA	N/a	L3 (caudal end)	TPA: <5th percentile per gender according to healthy donors; ♀ 380 mm^2 ♂ 800 mm^2	531 $mm^{2\ddagger}$ ♀ 423 $mm^{2\ddagger}$ ♂ 761 $mm^{2\ddagger}$	47%; ♀ 36% ♂ 58%
Meza-Junco, 2013	CSA (SMI)	SliceOmatic	L3	BMI ≥ 25: ♀ 41.0 cm^2/m^2 ♂ 53.0 cm^2/m^2 BMI < 25: 43.0 cm^2/m^2	54.0 $cm^2/m^{2\dagger}$	30%; ♀ 28% ♂ 31%
Montano-Loza, 2012	CSA (SMI)	SliceOmatic	L3	♀ 38.5 cm^2/m^2 ♂ 52.4 cm^2/m^2	51.0 $cm^2/m^{2\dagger}$	40%; ♀ 18% ♂ 50%

Author, Year	Muscles measured	Software	Level	Cut off values / definition	Muscle area / density	Sarcopenia prevalence
Montano-Loza, 2014	CSA (SMI)	SliceOmatic	L3	BMI ≥ 25: ♀ 41.0 cm^2/m^2 ♂ 53.0 cm^2/m^2 BMI < 25: 43.0 cm^2/m^2	50.0 $cm^2/m^{2\dagger}$	45% ♀ 30% ♂ 52%
Tandon, 2012	CSA (SMI)	SliceOmatic	L3	♀ 38.5 cm^2/m^2 ♂ 52.4 cm^2/m^2	♀ 44.9 $cm^2/m^{2\ddagger}$ ♂ 50.8 $cm^2/m^{2\ddagger}$	41%; ♀ 21% ♂ 54%
Toshima, 2015	TPA	N/a	L3 (caudal end)	TPA: <5th percentile per gender according to healthy donors; ♀ 380 $mm^{2¥}$ ♂ 800 $mm^{2¥}$	S: 484.2 $mm^{2\dagger¥}$ NS: 689.6 $mm^{2\dagger¥}$	46% ♀ 30% ♂ 52%
Tsien, 2014	TPA (PMI)	Leonardo Workstation using Oncocare	L4	PMI < 50 years: ♀ 10.5 cm^2/m^2 ♂ 12.3 cm^2/m^2 PMI > 50 years: ♀ 10.3 cm^2/m^2 ♂ 10.1 cm^2/m^2	PMI: 9.2 $cm^2/m^{2\dagger}$ (33.0 $HU^{\dagger}$)	62%
Valero, 2015	TPA, TPV	ImageJ, AW Workstation Volume	L3	PMI: ♀ 64.2 cm^2/m^2 ♂ 78.4 cm^2/m^2 TPV: ♀ 23.0 cm^3/m ♂ 34.1 cm^3/m	TPA¶: 784.4 $mm^2/m^{2\dagger}$ TPV: 30.4 $cm^3/m^{\dagger}$	49%
Waits, 2014	Morphometric age (including TPA, PD)	MATLAB	L4	Morphometric age (TPA, PD and AA calcification) as continuous variable	TPA: ♀ 14.6 $cm^{2\dagger}$ ♂ 23.1 $cm^{2\dagger}$ PD: ♀ 49.1 $HU^{\dagger}$ ♂ 48.8 $HU^{\dagger}$	n/a
Yadav, 2015	CSA (SMI)	SliceOmatic	L3	♀ 38.5 cm^2/m^2 ♂ 52.4 cm^2/m^2	54.3 $cm^2/m^{2\dagger}$ SMI survivors (alive/LT) 54.6 vs non-survivors 53.1 (deceased on waiting list), p=0.40	22% ♀ 13.1% ♂ 28.1% Sarcopenia survivors (alive/LT) 22% vs non-survivors (deceased on waiting list) 24%, p=0.77

† mean. ‡ median. * Any muscle contained within the region posterior to the spine and ribs, and no more lateral than the lateral-most edges of the erector spinae muscles. ¶ 743.1 mm^2/m^2 for liver transplant patients. ¥ Reported as cm^2 in the original article. Abbreviations: CSA; Cross Sectional Area, APMT; Axial Psoas Muscle Thickness; TPMT; Transversal Psoas Muscle Thickness, TPA; Total Psoas Area, IMAC; Intramuscular Adipose Content (defined as region of interest of multifidus muscle (Hounsfield units) divided by region of interest of subcutaneous fat (Hounsfield units)), TPV; Total Psoas Volume, PD; Psoas Density, N/a; Not available, L3; third lumbar vertebra; L4; fourth lumbar vertebra, T12; twelfth thoracic vertebra, HU; Hounsfield units, SMI; Skeletal Muscle Index (cm^2/m^2); PMI; Psoas Muscle Index (cm^2/m^2), BMI; Body Mass Index; AA; abdominal aneurysm, PSMA; Paraspinal Muscle Area, PSMI; Paraspinal Muscle Index, AWMA; Abdominal Wall Muscle Area, AWMI, Abdominal Wall Muscle Index.

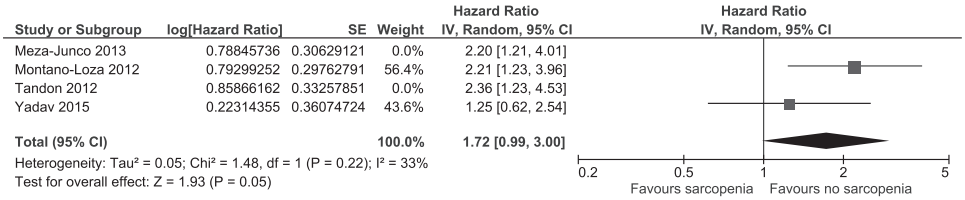
Table 3. Studies reporting the impact of sarcopenia on waiting list mortality in patients evaluated for liver transplantation or registered on the waiting list.

Author, Year	Survival
Durand, 2014	Pre-MELD: HR 0.92 (0.86-0.98), p=0.02 MELD-era: HR 0.86 (0.78-0.94), p=0.001 (for increasing TPMI)
Giusto, 2015	HR 0.89 (0.79-1.00)*
Meza-Junco, 2013	Median survival: S 16 (95% CI 4-28) vs NS 28 (21-34) months, log rank p=0.003 6-month survival S vs NS: 67% vs 90% 1-year survival S vs NS: 52% vs 82% Sarcopenia (with MELD/CP): HR 2.20 (1.21-4.02), p=0.01 Sarcopenia (individual components MELD/CP): 2.53 (1.35-4.73), p=0.004
Montano-Loza, 2012	Median survival: S 19 (7-30) vs NS 34 (14-55), log rank p=0.005 6-month survival S vs NS: 71% vs 90% 1-year survival S vs NS: 53% vs 83% Sepsis related death S vs NS: 22% vs 8%, p=0.02 Sarcopenia (with MELD/CP): HR 2.21 (1.23-3.95), p=0.008# Sarcopenia (individual components MELD/CP): 2.11 (1.13-3.94), p=0.02
Tandon, 2012	1-, 2-, and 5-year survival rates S vs NS: (63%, 51%, 51% vs 79%, 74%, 70% respectively), log-rank p=0.04; Low MELD (<15): log rank p=0.02; High MELD (≥15): log rank p=0.59 Sarcopenia: HR 2.36 (1.23-4.53), p=0.009
Yadav, 2015	Sarcopenia: HR 1.25 (0.62-2.55), p=0.54#

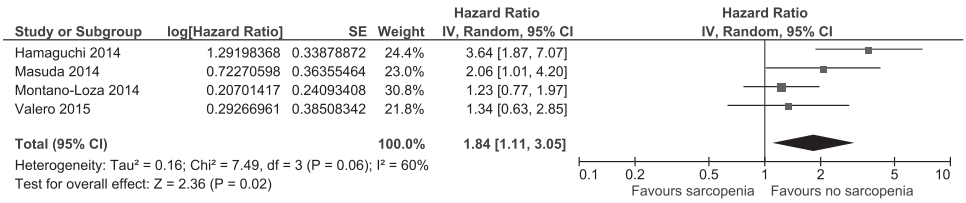
Unadjusted data. * Provided by the authors after personal communication. Abbreviations: HR; Hazard ratio, S; sarcopenic patients, NS; non-sarcopenic patients, SMI; Skeletal Muscle Index, TPMI; Transversal Psoas Muscle Index, TPA; Total Psoas Area, IMAC; Intramuscular Adipose Content, PMI; Psoas Muscle Index, DMG; Dorsal Muscle Group, MELD; Model for End-stage Liver Disease, CP; Child Pugh score, CI; confidence interval, MA; Muscle Attenuation, LT; Liver Transplantation.

Sarcopenia was also an independent predictor of mortality in patients evaluated for liver transplantation in the studies of Meza-Junco *et al.* and Montano-Loza *et al.* [23]. In both studies multivariable analyses were performed with MELD and Child Pugh scores on the one hand and with their individual components on the other hand, which all showed sarcopenia to be an independent predictor for mortality. Furthermore, Montano-Loza *et al.* reported a significantly higher sepsis related death in sarcopenic patients compared with non-sarcopenic patients (22% versus 8%, p=0.02), whereas no difference was found in liver failure related death [24]. Meza-Junco *et al.* reported a trend for higher liver failure related death in sarcopenic patients compared with non-sarcopenic patients (33% versus 15%, p=0.08 [23].

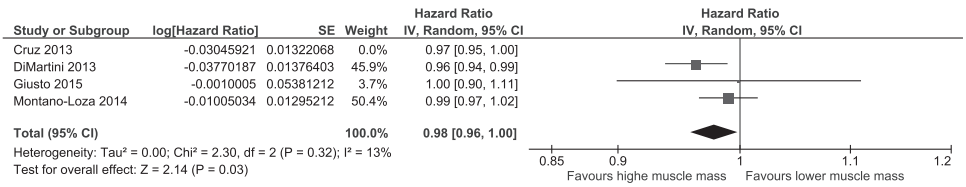
Figure 2. Forest plots of the association between sarcopenia and survival.



2a. Forest plot showing studies that reported the association between sarcopenia and waiting list mortality. Due to data provided by authors that was more precise than the data published in the article or rounding off upwards of downwards by Review Manager, the confidence intervals can somewhat differ from the original confidence intervals. For the study of Yadav *et al.* , unadjusted results were used because the multivariable analysis in the manuscript suggested a level of precision that did not correspond with the number of observed events. Because the studies of Meza-Junco *et al.* , Montano-Loza *et al.* , and Tandon *et al.* were performed in overlapping cohorts and the first was performed in patients with HCC, only the most representative study was included in the meta-analysis (i.e. all consecutive patients with cirrhosis being evaluated for liver transplantation) . The authors of these studies stated that at most fifteen patients were included in the study of Tandon *et al.* that were also included in the other studies. Including the study of Tandon *et al.* in the meta-analysis, resulted in a pooled HR of 1.93 (95% CI 1.33-2.80, p=0.0005), Z of 3.48 and I^2 of 1%.



2b. Forest plot showing studies that reported the association between sarcopenia and post-transplantation survival. Due to data provided by authors that was more precise than the data published in the article or rounding off upwards of downwards by Review Manager, the confidence intervals can somewhat differ from the original confidence intervals. Hamaguchi *et al.* , Masuda *et al.* , and Valero *et al.* performed measurements of the psoas muscle area, whereas Montano-Loza *et al.* performed measurements of the cross-sectional muscle area. A meta-analysis of studies that assessed skeletal muscle mass by measuring the psoas muscle area only resulted in a pooled HR of 2.21 (95% CI 1.25-3.90, p=0.007), Z of 2.72, and I^2 of 49%. A meta-analysis excluding the study of Valero *et al.* , that included only few patients that underwent liver transplantation for hepatocellular- or cholangiocarcinoma, resulted in a pooled HR of 2.04 (95% CI 1.05-3.92, p=0.03), Z of 2.11, and I^2 of 71% and a meta-analysis of Hamaguchi *et al.* and Masuda *et al.* resulted in a pooled HR of 2.78 (95% CI 1.59-4.85, p=0.0003), Z of 3.60 and I^2 of 24%.



2c. Forest plot showing studies that reported the association between skeletal muscle mass and post-transplantation survival. Only studies that reported the skeletal muscle index (cm^2/m^2) were included, as the other studies used different units of measurement. Due to data provided by authors that was more precise than the data published in the article or rounding off upwards of downwards by Review Manager, the confidence intervals can somewhat differ from the original confidence intervals. The hazard ratios shown represent an incremental increase in skeletal muscle index. Since DiMartini *et al.* and Cruz *et al.* performed studies in overlapping cohorts, only the latter was included in the meta-analysis. Additional results without stratification by gender that were provided by the authors of DiMartini *et al.* and these results were used in the meta-analysis. The authors also provided the results used for the study of Giusto *et al.* after personal communication .

Yadav *et al.* investigated the relationship between sarcopenia, six-minute walk distance and health-related quality of life in liver transplant candidates and found no association between sarcopenia and overall mortality. The unadjusted HR was 1.25 (95% CI 0.62-2.55, p=0.54) and was used for the meta-analysis rather than the adjusted HR, as the multivariable analysis suggested a level of precision that did not correspond with the number of observed events [30]. Although the mean MELD-scores were comparable between these studies, the MELD-score of the study cohort of Yadav *et al.* varied from 9 to 40 [30]. In the study of Giusto *et al.*, CT-assessed muscle mass was compared with Dual-Energy X-Ray Absorptiometry (DEXA) and anthropometry. The skeletal muscle index was not predictive for mortality on the waiting list (HR 0.89 [95% CI 0.79-1.00], kindly provided by the authors after personal communication), whereas mid-arm muscle circumference and fat-free mass index were, also after adjusting for sex, MELD-score, age, and interaction between sex and mid-arm muscle circumference and fat-free mass index, respectively [18]. However, the aim of this study was not to investigate the association between skeletal muscle mass and patient outcome and only 59 patients were included.

Post-transplantation survival

In the eleven studies that investigated the association between skeletal muscle mass and post-transplantation survival, seven described an association [14, 15, 17, 19, 21, 22, 29], and three no association [18, 25, 27, 28]. All details about median survival times, yearly survival rates and the association between skeletal muscle mass and overall survival are

summarized in table 4. The forest plot in figure 2b shows the association between sarcopenia and post-transplantation survival (pooled HR 1.84 [95% CI 1.11-3.05], p=0.02) with moderate heterogeneity between studies (I^2=60%). When studies that measured psoas muscle area were included only, this resulted in low heterogeneity (I^2=49%) and a pooled HR of 2.21 (95% CI 1.25-3.90, p=0.007). When the study of Valero *et al.* [28], that included only few patients that underwent liver transplantation for hepatocellular- or cholangiocarcinoma, was excluded, the pooled HR was 2.03 (95% CI 1.05-3.92, p=0.03, Z=2.11, I^2=71%). Finally, a meta-analysis of the studies of Hamaguchi *et al.* [19] and Masuda *et al.* [22] resulted in a pooled HR of 2.78 (95% CI 1.59-4.85, p=0.0003), Z of 3.60 and I^2 of 24%.

Meta-analyzing studies that reported the association between skeletal muscle index, as a continuous measure, and post-transplantation survival showed a pooled HR of 0.98 (95% CI 0.95-1.00, p=0.03) per incremental skeletal muscle index (figure 2c). Since DiMartini *et al.* [15] and Cruz *et al.* [14] performed studies in overlapping cohorts, only the latter was included in the meta-analysis. Additional results without stratification by gender were kindly provided by the authors of DiMartini *et al.* [15], and these results were used in the meta-analysis. The results used for the study of Giusto *et al.* were also kindly provided by the authors after personal communication [18].

Cruz *et al.* reported a protective effect of increasing skeletal muscle index on mortality (HR 0.97 [95% CI 0.94-0.99], p=0.04) [14], whereas the protective effect of the psoas muscle index was only found significant for males (HR 0.95, p=0.01) and not for females (HR 0.98, p=0.55) in the study of DiMartini *et al.* [15]. Every standard deviation increase in dorsal muscle group area, as assessed by Lee *et al.*, was also independently associated with increased overall (odds ratio [OR] 0.62 [95% 0.49-0.77], p<0.001), one-year (OR 0.53 [95% CI 0.36-0.78], p=0.001), and five-year (OR 0.53 [95% CI 0.38-0.70], p<0.001) survival, as well as total psoas area for one-year survival (OR 0.43 [95% CI 0.30-0.62], p<0.001) [21]. In line with this, Englesbe *et al.* also found an independent association between total psoas area and survival (HR 0.27 [0.14-0.53], p<0.001 per increasing 1000 mm^2) [17]. The variously defined parameter sarcopenia was an independent predictor for mortality in the study of Masuda *et al.* (HR 2.06 [95% CI 1.01-4.20], p=0.047) [22], high intramuscular adipose content (OR 3.90 [95% CI 2.03-7.76], p<0.001) and low PMI (OR 3.64 [1.90-7.17], p<0.001) have also been identified as independent predictors for impaired survival [19]. Waits *et al.* showed that morphometric age (including total psoas area, psoas density and abdominal aortic calcifications) was a risk factor for mortality per year increase (HR 1.03 [95% CI 1.02-1.04], p<0.001) [29].

Table 4. Studies reporting the impact of sarcopenia on overall survival in patients undergoing liver transplantation.

Author, Year	Survival
Cruz, 2013	SMI: HR 0.97 (0.94 – 0.99), p=0.04
DiMartini, 2013	♂ PMI: HR 0.95 (p=0.01) ♀ PMI: HR 0.98 (p=0.55)
Englesbe, 2010	1-year survival S vs NS: 49.7% vs 87% 3-year survival S vs NS: 26.4% vs 77.2% (lowest vs highest tertile) TPA: HR 0.27 (0.14-0.53), p<0.0001 (per increasing 1000 mm^2)
Giusto, 2015	HR 0.99 (0.90-1.11)*
Hamaguchi, 2014	Median survival low vs normal TPA: 17.6 vs 33.9 months Median survival high vs normal IMAC: 21.9 vs 32.4 months High IMAC: OR 3.90 (2.03-7.76), p<0.001 Low PMI: OR 3.64 (1.90-7.17), p<0.001
Lee, 2014	Overall survival: DMG: OR 0.62 (0.49-0.77), p<0.001 (per SD increase) 1-year survival: DMG: OR 0.53 (0.36-0.78), p=0.001 (per SD increase) TPA: OR 0.43 (0.30-0.62), p<0.001 (per SD increase) 5-year survival: DMG: OR 0.53 (0.38-0.70) p<0.001 (per SD increase)
Masuda, 2014	Sarcopenia: HR 2.06 (1.01-4.20), p=0.047 3-year survival S vs NS: 74.5% vs 88.9% (p=0.02) 5-year survival S vs NS: 69.7% vs 85.4% (p=0.02)
Montano-Loza, 2014	Median survival S vs NS: 117 (95% CI 84-151) vs 146 (95% CI 110-182) months, log rank p=0.4 1-year survival rate S vs NS: 89% vs 91% 5-year survival rate S vs NS: 74% vs 76% Sarcopenia: HR 1.23 (0.77-1.98), p=0.4[#] SMI: HR 0.99 (0.96-1.01), p=0.3[#] MA: HR 0.99 (0.96-1.02), p=0.5[#]
Tsien, 2014	Pre-OLT sarcopenia associated with mortality (p=0.06)[#] Non-significant association of continued reduction in muscle area with higher mortality (p=0.08)[#]
Valero, 2015	Median survival S vs NS: 38.5 vs 69.1 months (p=0.32) 1-year survival rate S vs NS: 76.6% vs 87.8% (p=0.15) 3-year survival rate S vs NS: 61.7% vs 71.4% (p=0.31) 5-year survival rate S vs NS: 55.3% vs 69.4% (p=0.32) Sarcopenia: HR 1.34 (0.61-2.76), p=0.43
Waits, 2014	Morphometric age: HR 1.03 (1.02-1.04), p<0.001 (per year) 1-year mortality morphometric age: OR 1.04 (1.03-1.06), p<0.001 (per year) 5-year mortality morphometric age: OR 1.03 (1.02-1.06), p<0.001 (per year)

[#] Unadjusted data. * Provided by the authors after personal communication. Abbreviations: HR; Hazard ratio, S; sarcopenic patients, NS; non-sarcopenic patients, SMI; Skeletal Muscle Index, TPMI; Transversal Psoas Muscle Index, TPA; Total Psoas Area, IMAC; Intramuscular Adipose Content, PMI; Psoas Muscle Index, DMG; Dorsal Muscle Group, MELD; Model for End-stage Liver Disease, CP; Child Pugh score, CI; confidence interval, MA; Muscle Attenuation, LT; Liver Transplantation.

Tsien *et al.* described a nonsignificant association between pretransplant sarcopenia and mortality (p=0.06) and higher mortality in patients with continued reduction in muscle area (p=0.08) in a relatively small cohort of 53 patients [27]. The median survival in the studies of Montano-Loza *et al.* [25] among cirrhosis patients undergoing liver transplantation and Valero *et al.* [28], among a relatively heterogeneous population of hepatocellular- and intrahepatic cholangiocarcinoma patients undergoing curative intent hepatic resection (70.9%) or liver transplantation (29.1%), did not significantly differ between sarcopenic and non-sarcopenic patients (117 versus 146 months, log rank p=0.4; and 38.5 versus 69.1 months, p=0.32, respectively). Neither sarcopenia [25, 28] nor skeletal muscle index or muscle attenuation [25] were predictive for mortality in regression models. Although Montano-Loza *et al.* found no association overall in a population with a relatively high MELD-score and hepatocellular carcinoma prevalence, male patients undergoing liver transplantation in the lowest skeletal muscle mass sextile showed significantly impaired survival compared with patients in the other sextiles [25]. Finally, Giusto *et al.* found no significant association between skeletal muscle index and post-transplant mortality after adjustment for age, gender and MELD-score (adjusted HR 1.0 [95% CI 0.90-1.11], data provided by the authors after personal communication) [18].

Post-transplantation complications and transplantation related mortality

In both studies reporting overall post-transplantation complications, low skeletal muscle mass was associated with increased risk of postoperative complications (table 5) [21, 28]. The study of Lee *et al.* showed that an increase in both the total psoas area and the dorsal muscle group with one standard deviation was associated with an increased risk for complications within one year after transplantation (OR 0.48 [95% CI 0.32-0.72], p<0.001, and OR 0.67 [95% CI 0.50-0.90], p=0.007, per standard deviation increase in dorsal muscle group area) [21]. In the study of Valero *et al.* post-transplantation complications occurred in 40.4% of sarcopenic patients compared with 18.4% in non-sarcopenic patients (p=0.01). Sarcopenia was an independent predictor for postoperative complications in multivariable analysis (OR 3.06 [95% CI 1.07-8.72], p=0.03) [28]. All severe postoperative (23.4%) complications (i.e. Clavien-Dindo classification ≥ IIIa) occurred in patients with sarcopenia. No differences were observed in 30- and 90-day mortality rates in sarcopenic and non-sarcopenic patients respectively (4.3% versus 0%, p=0.24 and 8.5% versus 2.0%, p=0.20) [28]. The three-month mortality rate in the study of Montano-Loza *et al.* was 5% in sarcopenic patients compared with 2% in non-sarcopenic patients (p=0.20) [25]. In the study of DiMartini *et al.*, the relative risk for in-hospital mortality was 0.97 (p>0.05) [15].

Table 5. Studies reporting the impact of sarcopenia on short-term outcome in patients who underwent liver transplantation.

Author, Year	Complications				Length of Stay		
	All	Clavien Dindo classification ≥ IIIa	Mortality	Infectious	ICU	Hospital (days)	Intubation time
DiMartini, 2013	N/a	N/a	In-hospital: ♀ RR 0.97 (N.s.) ♂ RR 0.97 (N.s.)	N/a	p<0.001	p<0.001 Discharge to hospital/ nursing home: ♀ RR 0.96 (p<0.05) ♂ RR 0.95 (N.s.)	Intubation days (p<0.001)
Krell, 2013	N/a	N/a	N/a	Any infection: Low TPA vs high TPA OR 4.6 (2.25-9.53) Severe infections: decreasing TPA HR 0.38 (0.23-0.65) (p<0.01)	N/a	N/a	N/a
Lee, 2014	Within 1 year TPA: OR 0.48 (0.32-0.72, p<0.001) per SD increase DMG: OR 0.67 (0.50-0.90, p=0.007) per SD increase	N/a	N/a	N/a	N/a	N/a	N/a
Masuda, 2014	N/a	N/a	N/a	Sepsis: Sarcopenia 17.7% vs. no sarcopenia 7.4%; HR 5.31 (1.53-18.4), p=0.009	N/a	N/a	N/a
Montano-Loza, 2014	N/a	N/a	3-month: S 5% vs NS 2%, p=0.2	Overall 90-day infections: S 29% vs NS 20%, p=0.1 Bacterial infections: S 26% vs NS 15%, p=0.04	S 12 days vs NS 6 days, p=0.001	S 40 days vs NS 25 days, p=0.005	N/a
Toshima, 2015	N/a	N/a	N/a	Sepsis: OR sarcopenia 1.72 (0.67-5.03), p=0.263	N/a	N/a	N/a
Tsien, 2014	Psoas and paraspinal muscle mass reduction OR for DM 3.1 (1.01-9.38), p<0.05	N/a	N/a	N/a	TPA: p>0.1	TPA: p>0.1	N/a
Valero, 2015	Sarcopenia 40.4% vs no sarcopenia 18.4%, p=0.01; OR sarcopenia 3.06 (1.07-8.72), p=0.03	All severe complications (23.4%) occurred in sarcopenic group.	30 day: S 4.3% vs NS 0% (p=0.24) 90 day: S 8.5% vs NS 2.0% (p=0.20)	N/a	N/a	S 12.1 vs NS 9.7 days, p=0.50	N/a

† mean. ‡ median. Abbreviations; ICU Intensive care unit, OR; Odds ratio, HR: Hazard ratio, N.s.; Not significant, TPA; Total Psoas Area; DMG; Dorsal Muscle Group, DM; Diabetes Mellitus, S; sarcopenic patients, NS; non-sarcopenic patients, N/a; Not available.

Krell *et al.* reported that patients with a total psoas area in the lowest tertile had a 4.6-fold increased risk (95% CI 2.25-9.53) to develop any post-transplantation infection compared with patients in the highest tertile. In a multivariable model, pretransplant total psoas area was an independent predictor for the occurrence of severe infections (HR 0.38 [95% CI 0.23-0.65], p<0.01) together with age and pretransplant bilirubin level. These factors remained significant when infections were stratified by pathogen type (i.e. bacterial, viral or fungal) [20]. In line with this, Masuda *et al.* found that sarcopenia was an independent predictor for sepsis (HR 5.31 [95% CI 1.53-18.40], p=0.009) in a cohort of 228 patients, which occurred in 17.7% of sarcopenic patients and 7.4% of non-sarcopenic patients [22]. On the other hand, no independent association was found between sarcopenia and sepsis in a sub study in the same patient cohort (n=143) of Toshima *et al.* (OR 1.72 [95% CI 0.67-5.03], p=0.263) [26]. However, both studies performed a multivariable analysis of risk factors for postoperative sepsis including ten and thirteen parameters respectively on only 25 (49%) and twelve events respectively. Therefore, the methodology of these studies could be questioned. Although the overall 90-day infection rate did not significantly differ between sarcopenic and non-sarcopenic patients (29% versus 20%, p=0.1) in a study of Montano-Loza *et al.*, bacterial infections in particular within 90 days after transplantation occurred significantly more in sarcopenic patients compared with non-sarcopenic patients (26% versus 15%, p=0.04) [25]. However, no multivariable analysis has been performed for bacterial infections.

Only one study reported that five patients had confirmed acute graft rejection without specifying them as (non-)sarcopenic [27].

Post-transplantation length of hospital stay

Four studies reported on length of stay outcomes (table 5) [15, 25, 27, 28]. DiMartini *et al.* performed a Poisson regression analysis and found skeletal muscle mass to be predictive for length of both hospital and ICU stay, as well as intubation days (all p<0.001) [15]. Furthermore, sarcopenic patients were more likely to be discharged to another hospital or nursing home rather than home (p=0.04) [15]. A significantly increased length of ICU (12 days versus 6 days, p=0.001) and hospital stay (40 days versus 25 days, p=0.005) was also found in cirrhosis patients undergoing liver transplantation by Montano-Loza *et al.* [25], whereas Valero *et al.* found non-significant differences regarding hospital stay between sarcopenic and non-sarcopenic patients who underwent hepatic resection with curative intent or transplantation for hepatocellular- or intrahepatic cholangiocarcinoma (12.1 days versus 9.7 days, p=0.50) [28]. Obviously, these patients have a distinct postoperative recovery compared with transplant patients. Furthermore, Tsien *et al.* [27], who measured the psoas area in only 53 patients, found no association with length of hospital stay in contrast to Montano-Loza *et al.* [25] and DiMartini *et al.* [15], who performed cross-sectional skeletal muscle measurements in larger cohorts.

DISCUSSION

In recent years, multiple narrative reviews regarding sarcopenia among transplantation patients have been published [31-33]. This is the first systematic review of studies that investigated the influence of skeletal muscle mass by means of abdominal CT in patients who were evaluated for or underwent liver transplantation or were registered on the waiting list. According to the current findings, there is consistent evidence that sarcopenia is associated with impaired survival, independent of other risk factors such as age and MELD-score or its individual components. This association was found both before and after the introduction of the MELD-score (i.e. in the United States in 2002) [34]. Due to a substantial heterogeneity between reported outcome measures, less consistent evidence suggests that sarcopenia is associated with post-transplantation complications, which may be infectious complications in particular.

Some findings of this systematic review are conflicting and multiple reasons could be postulated. Definitions of sarcopenia greatly varied, as there currently is no consensus regarding adequate cut-off values. Although liver transplant patients greatly differ from cancer patients, most studies used cut-off values based on oncological studies, such as those defined by Prado and colleagues [35]. This could have led to inadequate classification of patients as (non-)sarcopenic. Therefore, one could wonder whether one set of cut-off values would be applicable for various populations and gender-, age-, ethnicity-, and disease-specific cut-off values may be needed. One study described the association between morphometric age, but the association between the individual skeletal muscle mass components (i.e. psoas area and psoas density) was not described [29]. Besides multiple definitions for sarcopenia, multiple methods to perform skeletal muscle mass measurements (e.g., psoas area, cross-sectional area) have been used throughout studies.

Sarcopenia was associated with waiting list mortality in four studies [16, 23, 24], whereas two other studies reported no significant association [18, 30]. Besides different cut-off values and methods used, an explanation of this difference could be a varying range of MELD-score within these studies. After all, the association between sarcopenia and waiting list mortality was found in patients with lower MELD-scores in particular [16, 24]. No association between sarcopenia and overall post-transplantation survival was reported in three studies [18, 25, 28]. The varying study populations could be an explanation for these conflicting results, as well as the use of BMI- and sex-specific cut-off values that are used to predict survival in cancer patients, in transplant populations [36]. Meza-Junco *et al.* included hepatocellular cancer patients who underwent liver transplantation only and used the same cut-off values. They indeed found an independent association

with survival [23]. In all but one study with questionable methodology [26], describing the association between pre-transplantation sarcopenia and post-transplantation short-term outcome, sarcopenia was independently associated with complications and mortality.

This systematic review postulates that skeletal muscle mass is a prognostic factor, independent of MELD-score. Although it remains to be investigated whether skeletal muscle mass assessment is superior to the 'eyeball test', i.e. the subjective clinician's assessment of a patient's physical status or frailty, it could objectively underscore subjective assessments [17]. The study of Tandon *et al.* showed that sarcopenic patients with a low MELD-score had a similar outcome compared with patients with a high MELD-score with or without sarcopenia [24]. Therefore, skeletal muscle mass assessment may be used to more accurately select liver transplant patients and allocate organs in the future. After all, a selection of patients that is at risk for early mortality is probably not adequately identified by the MELD-score, since 71% of the patients who died on the waiting list had a MELD-score ≤25 at registration in the study of Durand *et al.* [16]. Although this would be a challenge due to the current organ shortage, patients with a low MELD-score and low skeletal muscle mass could, for example, be prioritized on the waiting list or be selected for targeted treatment of muscle wasting. Currently, such trials are being performed [37] and potential drugs are being investigated [38] in cancer populations.

Despite the strong prognostic value of the MELD-score, the survival of 15-20% of the patients cannot accurately be predicted [34]. The most frequently reported limitation of the MELD-score is the lack of parameters reflecting patients' nutritional and functional status. Therefore, modifications of the MELD-score, such as the MELDNa-score and the 5-variable MELD-score, including serum sodium and albumin levels respectively, have been developed [4, 5, 39]. Both scores improved mortality prediction. The outcome of this systematic review supports that sarcopenic liver transplant candidates face a worsened outcome. Besides the superiority of the MELD-psoas area score of Durand *et al.* over the MELD-score and MELDNa-score to predict waiting list mortality [16], the predictive value of the MELD-Sarcopenia score was found superior compared with the MELD-score in a recent study [40]. However, validation of these scores is recommended. If validated, clinical trials are warranted to investigate whether transplantation in sarcopenic patients with lower MELD-scores may be preferential. Prospective trials are also needed to investigate the natural course of sarcopenia following liver transplantation. Little is known on whether skeletal muscle mass fully normalizes, and how post-transplantation change in muscle mass impacts outcome, such as the development of post-transplantation diabetes mellitus [27].

Preoperative risk assessment remains of paramount importance in patients who have been allocated a donor liver. Some studies suggest that a poor nutritional status, for instance reflected by the subjective global assessment, impairs post-transplantation outcomes [41-43]. However, these measures are frequently considered as subjective [44]. Furthermore, limiting factors, such as fluid retention, could hamper nutritional assessment [16]. Single-slice CT-assessed skeletal muscle measurements are considered an objective and easy-to-perform method with high inter-observer agreement that could be performed on routinely available CT scans [8]. Up to now, no gold standard has been established to perform body composition measurements. However, CT imaging was the method of preference in an expert consensus meeting on cachexia [45], particularly in cirrhotic patients who frequently have ascites [18].

Some limitations of the current review and included studies should be mentioned. First, all included studies were retrospective, observational cohort studies. Although this may have resulted in selection bias, the study cohorts consisted of non-selected, consecutive patients listed for or undergoing liver transplantation. Second, selective publication of data could have led to an underestimation of negative results. Third, four research groups from Canada, [23-25], Ann Arbor [17, 20, 21, 29] and Pittsburg [13-15], USA, and Japan [22, 26] published multiple original articles including patient populations from their center that were all included in the current review. These four centers contributed to thirteen of the nineteen studies. Therefore, the number of patient cohorts studied is likely to be smaller than the number of original articles included in this systematic review.

In conclusion, sarcopenia impairs outcome in patients awaiting or undergoing liver transplantation. Skeletal muscle mass assessment may contribute to pre-transplantation risk assessment.

REFERENCES

1. Edge SB BD, Compton CC, Fritz AG, Greene FL, Trotti A, editors. AJCC cancer staging manual (7th ed). New York, NY: Springer; 2010.
2. Freeman RB, Jr., Wiesner RH, Harper A, McDiarmid SV, Lake J, Edwards E, et al. The new liver allocation system: moving toward evidence-based transplantation policy. Liver Transpl 2002; 8: 851-8.
3. Brown RS, Rush SH, Rosen HR, Langnas AN, Klintmalm GB, Hanto DW, et al. Liver and intestine transplantation. Am J Transplant 2004; 4 Suppl 9: 81-92.
4. Myers RP, Shaheen AA, Faris P, Aspinall AI, Burak KW. Revision of MELD to include serum albumin improves prediction of mortality on the liver transplant waiting list. PLoS One 2013; 8: e51926.
5. Myers RP, Tandon P, Ney M, Meeberg G, Faris P, Shaheen AA, et al. Validation of the five-variable Model for End-stage Liver Disease (5vMELD) for prediction of mortality on the liver transplant waiting list. Liver Int 2014; 34: 1176-83.
6. Lai JC, Feng S, Terrault NA, Lizaola B, Hayssen H, Covinsky K. Frailty predicts waitlist mortality in liver transplant candidates. Am J Transplant 2014; 14: 1870-9.
7. Levolger S, van Vugt JL, de Bruin RW, IJzermans JN. Systematic review of sarcopenia in patients operated forresectable gastrointestinal and hepatopancreatobiliary malignancies. Br J Surg 2015; 102: 1448-58.
8. Reisinger KW, van Vugt JL, Tegels JJ, Snijders C, Hulsewe KW, Hoofwijk AG, et al. Functional compromise reflected by sarcopenia, frailty, and nutritional depletion predicts adverse postoperative outcome after colorectal cancer surgery. Annals of surgery 2015; 261: 345-52.
9. van Vugt JL, Braam HJ, van Oudheusden TR, Vestering A, Bollen TL, Wiezer MJ, et al. Skeletal Muscle Depletion is Associated with Severe Postoperative Complications in Patients Undergoing Cytoreductive Surgery with Hyperthermic Intraperitoneal Chemotherapy for Peritoneal Carcinomatosis of Colorectal Cancer. Ann Surg Oncol 2015; 22: 3625-31.
10. Moher D, Liberati A, Tetzlaff J, Altman DG, Group P. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. Int J Surg 2010; 8: 336-41.
11. Wells GA, Shea B, O'connell D, Peterson JEA, Welch V, Losos M, et al. The Newcastle-Ottawa Scale (NOS) for assessing the quality of nonrandomised studies in meta-analyses. [cited 2015 20 May]; Available from: http://www.ohri.ca/programs/clinical_epidemiology/oxford.asp.
12. Higgins JP, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. Bmj 2003; 327: 557-60.
13. Bergerson JT, Lee JG, Furlan A, Sourianarayanane A, Fetzer DT, Tevar AD, et al. Liver transplantation arrests and reverses muscle wasting. Clin Transplant 2014.

14. Cruz RJ, Jr., Dew MA, Myaskovsky L, Goodpaster B, Fox K, Fontes P, et al. Objective radiologic assessment of body composition in patients with end-stage liver disease: going beyond the BMI. Transplantation 2013; 95: 617-22.
15. DiMartini A, Cruz Jr RJ, Dew MA, Myaskovsky L, Goodpaster B, Fox K, et al. Muscle mass predicts outcomes following liver transplantation. Liver Transplant 2013; 19: 1172-80.
16. Durand F, Buyse S, Francoz C, Laouenan C, Bruno O, Belghiti J, et al. Prognostic value of muscle atrophy in cirrhosis using psoas muscle thickness on computed tomography. J Hepatol 2014; 60: 1151-7.
17. Englesbe MJ, Patel SP, He K, Lynch RJ, Schaubel DE, Harbaugh C, et al. Sarcopenia and mortality after liver transplantation. Journal of the American College of Surgeons 2010; 211: 271-8.
18. Giusto M, Lattanzi B, Albanese C, Galtieri A, Farcomeni A, Giannelli V, et al. Sarcopenia in liver cirrhosis: the role of computed tomography scan for the assessment of muscle mass compared with dual-energy X-ray absorptiometry and anthropometry. Eur J Gastroenterol Hepatol 2015; 3: 328-34.
19. Hamaguchi Y, Kaido T, Okumura S, Fujimoto Y, Ogawa K, Mori A, et al. Impact of quality as well as quantity of skeletal muscle on outcomes after liver transplantation. Liver Transpl 2014; 20: 1413-9.
20. Krell RW, Kaul DR, Martin AR, Englesbe MJ, Sonnenday CJ, Cai S, et al. Association between sarcopenia and the risk of serious infection among adults undergoing liver transplantation. Liver Transpl 2013; 19: 1396-402.
21. Lee CS, Cron DC, Terjimanian MN, Canvasser LD, Mazurek AA, Vonfoerster E, et al. Dorsal muscle group area and surgical outcomes in liver transplantation. Clin Transplant 2014; 28: 1092-8.
22. Masuda T, Shirabe K, Ikegami T, Harimoto N, Yoshizumi T, Soejima Y, et al. Sarcopenia is a prognostic factor in living donor liver transplantation. Liver Transpl 2014; 20: 401-7.
23. Meza-Junco J, Montano-Loza AJ, Baracos VE, Prado CM, Bain VG, Beaumont C, et al. Sarcopenia as a prognostic index of nutritional status in concurrent cirrhosis and hepatocellular carcinoma. J Clin Gastroenterol 2013; 47: 861-70.
24. Tandon P, Ney M, Irwin I, Ma MM, Gramlich L, Bain VG, et al. Severe muscle depletion in patients on the liver transplant wait list: its prevalence and independent prognostic value. Liver Transpl 2012; 18: 1209-16.
25. Montano-Loza AJ, Meza-Junco J, Baracos VE, Prado CM, Ma M, Meeberg G, et al. Severe muscle depletion predicts postoperative length of stay but is not associated with survival after liver transplantation. Liver Transpl 2014; 20: 640-8.
26. Toshima T, Shirabe K, Kurihara T, Itoh S, Harimoto N, Ikegami T, et al. Profile of plasma amino acids values as a predictor of sepsis in patients following living donor liver transplantation: Special reference to sarcopenia and postoperative early nutrition. Hepatol Res 2015; [Epub ahead of print].

27. Tsien C, Garber A, Narayanan A, Shah SN, Barnes D, Eghtesad B, et al. Post-liver transplantation sarcopenia in cirrhosis: a prospective evaluation. J Gastroenterol Hepatol 2014; 29: 1250-7.

28. Valero V, 3rd, Amini N, Spolverato G, Weiss MJ, Hirose K, Dagher NN, et al. Sarcopenia adversely impacts postoperative complications following resection or transplantation in patients with primary liver tumors. Journal of gastrointestinal surgery : official journal of the Society for Surgery of the Alimentary Tract 2015; 19: 272-81.

29. Waits SA, Kim EK, Terjimanian MN, Tishberg LM, Harbaugh CM, Sheetz KH, et al. Morphometric age and mortality after liver transplant. JAMA Surg 2014; 149: 335-40.

30. Yadav A, Chang YH, Carpenter S, Silva AC, Rakela J, Aqel BA, et al. Relationship between sarcopenia, six-minute walk distance and health-related quality of life in liver transplant candidates. Clin Transplant 2015; 29: 134-41.

31. Carey EJ. Sarcopenia in solid organ transplantation. Nutr Clin Pract 2014; 29: 159-70.

32. Kim HY, Jang JW. Sarcopenia in the prognosis of cirrhosis: Going beyond the MELD score. World J Gastroenterol 2015; 21: 7637-47.

33. Montano-Loza AJ. Muscle wasting: a nutritional criterion to prioritize patients for liver transplantation. Current opinion in clinical nutrition and metabolic care 2014; 17: 219-25.

34. Kamath PS, Kim WR, Advanced Liver Disease Study G. The model for end-stage liver disease (MELD). Hepatology 2007; 45: 797-805.

35. Prado CM, Lieffers JR, McCargar LJ, Reiman T, Sawyer MB, Martin L, et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. Lancet Oncol 2008; 9: 629-35.

36. Martin L, Birdsell L, Macdonald N, Reiman T, Clandinin MT, McCargar LJ, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol 2013; 31: 1539-47.

37. Reisinger KW, Bosmans JWAM, Uittenbogaart M, Alsoumali A, Poeze M, Sosef MN, et al. Loss of Skeletal Muscle Mass During Neoadjuvant Chemoradiotherapy Predicts Postoperative Mortality in Esophageal Cancer Surgery. Ann Surg Oncol 2015; 22: 4445-52.

38. Dingemans AM, de Vos-Geelen J, Langen R, Schols AM. Phase II drugs that are currently in development for the treatment of cachexia. Expert Opin Investig Drugs 2014; 23: 1655-69.

39. Kim WR, Biggins SW, Kremers WK, Wiesner RH, Kamath PS, Benson JT, et al. Hyponatremia and mortality among patients on the liver-transplant waiting list. The New England journal of medicine 2008; 359: 1018-26.

40. Montano-Loza AJ, Duarte-Rojo A, Meza-Junco J, Baracos VE, Sawyer MB, Pang JX, et al. Inclusion of Sarcopenia Within MELD (MELD-Sarcopenia) and the Prediction of Mortality in Patients With Cirrhosis. Clinical and translational gastroenterology 2015; 6: e102.

41. Merli M, Giusto M, Gentili F, Novelli G, Ferretti G, Riggio O, et al. Nutritional status: its influence on the outcome of patients undergoing liver transplantation. Liver Int 2010; 30: 208-14.

42. Selberg O, Bottcher J, Tusch G, Pichlmayr R, Henkel E, Muller MJ. Identification of high- and low-risk patients before liver transplantation: a prospective cohort study of nutritional and metabolic parameters in 150 patients. Hepatology 1997; 25: 652-7.

43. Stephenson GR, Moretti EW, El-Moalem H, Clavien PA, Tuttle-Newhall JE. Malnutrition in liver transplant patients: preoperative subjective global assessment is predictive of outcome after liver transplantation. Transplantation 2001; 72: 666-70.

44. Englesbe MJ. Quantifying the eyeball test: sarcopenia, analytic morphomics, and liver transplantation. Liver Transpl 2012; 18: 1136-7.

45. Fearon K, Strasser F, Anker SD, Bosaeus I, Bruera E, Fainsinger RL, et al. Definition and classification of cancer cachexia: an international consensus. Lancet Oncol 2011; 12: 489-95.

CHAPTER 11

A Nomogram with Sarcopenia Surpasses the MELD Score in Predicting Waiting List Mortality in Cirrhotic Liver Transplant Candidates: A Competing Risk Analysis in a National Cohort

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ABSTRACT

Background: Frail patients with low MELD scores may be under prioritised. Low skeletal muscle mass (i.e. sarcopenia) has been identified as risk factor for waiting list mortality and a recent study proposed to incorporate sarcopenia in the MELD score (i.e. MELD-Sarcopenia score). We aimed to investigate the association between sarcopenia and waiting list mortality, and to validate the MELD-Sarcopenia score (i.e. MELD+10.35*Sarcopenia).

Methods: We identified consecutive patients with cirrhosis listed for liver transplantation in the Eurotransplant registry between 2007-2014 and mesures skeletal muscle mass on computed tomography (CT). A competing risk analysis was used to compare survival of patients with and without sarcopenia, and concordance (c) indices were used to assess performance of the MELD and MELD-Sarcopenia score. We created a nomogram of the best predictive model.

Results: We included 585 patients with a median MELD of 14 (IQR 9-19), of which 254 (43.4%) were identified as having sarcopenia. Median waiting list survival was shorter in patients with sarcopenia than those without ($p<0.001$). This effect was even more pronounced in patients with MELD ≤15. The discriminative performance of the MELD-Sarcopenia score (c-index 0.820) for 3-month mortality was lower than MELD score alone (c-index 0.839). Apart from sarcopenia and MELD score, other predictive variables were occurrence of hepatic encephalopathy before listing, and recipient age. A model including all these variables yielded a c-index of 0.851.

Conclusions: Sarcopenia was associated with waiting list mortality in liver transplant candidates with cirrhosis, particularly in patients with lower MELD scores. The MELD-Sarcopenia score was successfully validated in this cohort. However, incorporating sarcopenia in the MELD score had limited added value in predicting waiting list mortality.

INTRODUCTION

Model for end-stage liver disease (MELD) score is the most frequently used method to prioritise patients with end-stage livers disease for liver transplantation and it is calculated using serum levels of bilirubin and creatinine and the international normalized ratio (INR) [1]. Despite its strong predictive value, disease severity is underestimated in about 15-20% pf cirrhotic patients by the MELD score, resulting in an inaccurate prediction of survival [2]. Amongst others, conditions such as hyponatremia and hypoalbuminemia have been identified as additional risk factors for impaired waiting list survival. This knowledge resulted in modifications of the original MELD score; i.e. the MELDNa and five-variable MELD score, respectively [3-5]. Moreover, a frequently reported drawback of the MELD score is the lack of an objective parameter reflecting patients' physical and nutritional status, as was albumin in the old Child-Turcotte-Pugh score. Consequently, patients with a biochemically low MELD score, but with malnutrition or low skeletal muscle mass (sarcopenia), may be under prioritised in the current system [6]. Indeed, sarcopenia, a hallmark sign of frailty and functional decline [7, 8], has recently been found to predict waiting list mortality [9, 10]. Montano-Loza *et al.* found a significantly shorter waiting list survival in patients with sarcopenia, and therefore included sarcopenia in the MELD score (i.e. MELD-Sarcopenia score). This score showed a higher predictive accuracy for waiting list mortality than the MELD score alone [10].

This particular MELD-Sarcopenia score is of great interest for the generally catabolic cirrhotic population. However, the MELD-Sarcopenia score has not been externally validated. And, in addition, most studies investigating the association between sarcopenia and waiting list mortality were performed in North-American populations [9-15], which differ from Western-European populations (e.g., regarding body mass index (BMI), race, and healthcare accessibility). Furthermore, they were hampered by limited sample size and methodology, while not taking into account competing risks on the waiting list [11-14, 16]. Particularly in transplant patients, competing risk analyses are considered superior to proportional hazard models [17, 18]. Therefore, our aims were 1) to investigate the association between sarcopenia and waiting list mortality in a Western-European cohort using competing risk analysis, 2) to validate the MELD-Sarcopenia score, and 3) to identify the best performing predictive model in patients with cirrhosis listed for liver transplantation.

MATERIALS AND METHODS

Patients and data acquisition

We identified all consecutive adult (≥18 years) patients with cirrhosis who have been placed on the waiting list for liver transplantation in one of the liver transplantation centers in the Netherlands (i.e. Erasmus MC University Medical Center, Rotterdam; Leiden University Medical Center, Leiden; and Groningen University Medical Center, Groningen) between 2007-2014, using the prospective Eurotransplant registry [19]. Patients listed for retransplantation, multivisceral transplantation, candidates on the high urgency list, or patients below 18 years old were excluded. All registry data was collected prospectively, except for the occurrence of liver-related complications before listing (i.e. ascites, hepatic encephalopathy, spontaneous bacterial peritonitis, esophageal variceal bleeding), which was collected retrospectively. Patients with hepatocellular carcinoma (HCC) were transplanted only if they fulfilled the Milan criteria [20]. The MELD score and recipient age were calculated at the time of listing. The study protocol conforms to the ethical guidelines of the 1975 Declaration of Helsinki and was approved by the Institutional Review Boards of all involved centers. A waiver for informed consent was granted.

Skeletal muscle mass measurements

Computed tomography (CT) scanning is the gold standard to measure skeletal muscle mass, particularly in end-stage liver disease patients, since these measurements are not influenced by the presence of ascites or edema [16]. For the purpose of this study we used CT examinations which were performed routinely as part of the liver transplant evaluation. Since liver transplant candidates are prone to skeletal muscle wasting over time, we included only those CT examinations that were performed within 90 days from waiting list placement. We calculated the cross-sectional skeletal muscle area (CSMA; figure 1) and the skeletal muscle index (SMI), as previously described in more detail [21]. In short, the CSMA was calculated using the transversal slice at the level of the third lumbar vertebra (L3 in cm^2) and divided by patients' squared height resulting in the skeletal muscle index (SMI, cm^2/m^2). We chose L3 as it has been shown that the SMI at the level of L3-L4 gives an accurate estimation of total body skeletal muscle mass [22]. A previously validated non-commercial software tool developed at the Erasmus MC University Medical Center was used for the calculation of the CSMA [23].

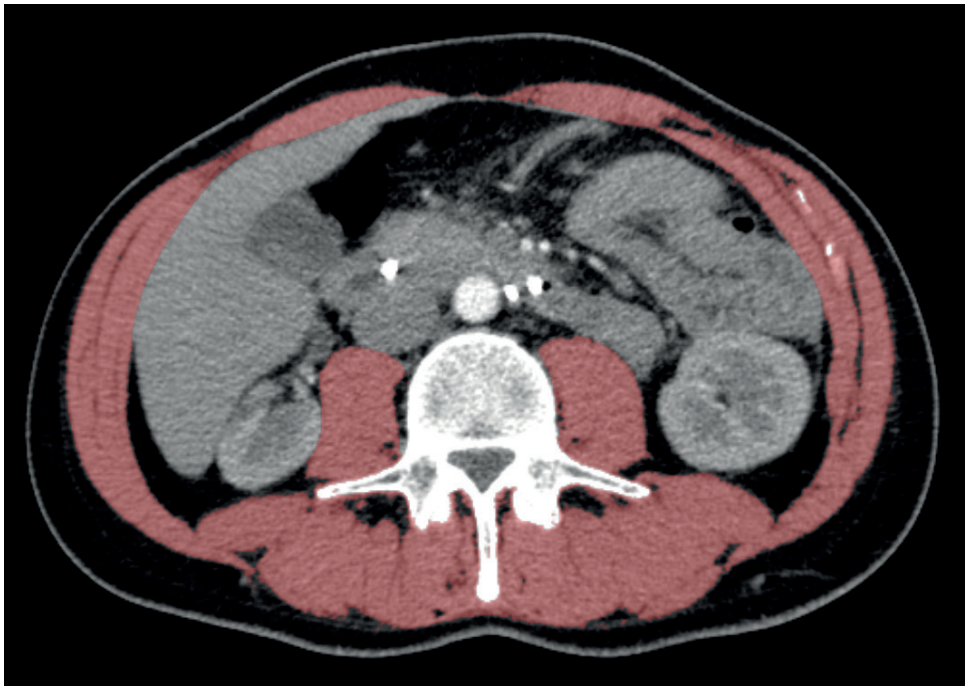

Figure 1. Cross-sectional skeletal muscle area (CSMA, in cm^2) at the level of the third lumbar vertebra (L3).

The MELD-Sarcopenia score

The MELD-Sarcopenia score, as established by Montano-Loza *et al.* [10], is based on pre-established cut-off values for the SMI (i.e. males with BMI <25: <43 cm^2/m^2, males with BMI ≥25: <53 cm^2/m^2, females: <41 cm^2/m^2) [24]. Therefore, we classified patients as having sarcopenia using the same cut-off values. As the MELD-Sarcopenia score was calculated as follows: MELD + (10.35 * Sarcopenia) [10], this corresponds with a 10.35-point increase in the MELD score in patients with sarcopenia. In addition, to test the robustness of our data, patients were also classified as having sarcopenia using another recently developed cut-off values in a cohort of end-stage liver disease patients by Carey *et al.*; 50 cm^2/m^2 for men and 39 cm^2/m^2 for women [25].

Statistical analyses

Categorical data are reported as counts with percentages. Continuous data are reported as the median with interquartile range (IQR). The Chi-square test was used to compare categorical data. Depending on the normality of the distribution (non)parametrical tests were used to compare continuous data. Multiple imputations were performed, using 5

imputed datasets to correct for bias due to missing values (i.e. serum albumin (n=73, 12.5%) and sodium levels (n=74, 12.6%) as they had not been routinely performed in one center). We used MICE package for R version 3..3.3 for this multiple imputation. Because these parameters were not associated with waiting list mortality after backward stepwise selection, we performed a complete-case analysis.

The primary endpoint of this study was waiting list mortality. To account for immortal time bias, waiting list time (i.e. survival) was defined from the date of CT onwards until transplantation, removal from the waiting list, or death occured. Patients who were removed from the waiting list because of clinical deterioration or progression of HCC outside of the Milan criteria were considered deceased at time of removal if they indeed died within 3 months after waiting list removal. We censored patients who were removed for reasons other than abovementioned, as well as patients who were still on the waiting list on 31 December 2016. Survival status was checked using the municipal record database.

We regarded the chance of occurrence of our primary endpoint and the chance of transplantation as competing risks. Therefore, we plotted the cumulative incidence functions for patients with and without sarcopenia according to the definition of Martin *et al.* (Sarcopenia$_M$) [24] and Carey *et al.* (Sarcopenia$_C$) [25] with transplantation as competing risk. Secondly, we assessed the performance of the MELD and MELDNa scores with or without adjustment for Sarcopenia$_M$ and Sarcopenia$_C$, and the MELD-Sarcopenia score by calculating the concordance index (c-index) using Wolbers' method and the subdistribution hazard as proposed by Fine and Gray.. The method by Wolbers *et al.* is an adaptation of Harrell's concordance index for competing risk analyses [26, 27]. The c-indices were calculated for 3-month waiting list survival, as the MELD score was designed to predict 3-month mortality, [2] and internal validation was performed using bootstrapping with 100 samples. The subdistribution hazard ratio (sHR) is the instantaneous risk of dying from a particular cause κ given that the subject has not died from cause κ. Finally, a multivariable competing risk analysis was performed, using a subdistribution hazards approach as proposed by Fine and Gray [17, 28], to identify risk factors for waiting list mortality. Using a backward stepwise selection based on the Akaike Information Criterion (AIC), factors were selected for the final model The highest performance (based on the c-index), age, serum albumin level, serum sodium level, and complications before listing were included in the full model before selection. As patients with a low MELD score and low skeletal muscle mass may benefit from a higher priority on the waiting list, a subgroup analysis was performed in patients with a MELD score ≤15. Each independent parameter associated with impaired survival was assigned a specific weighted score

using the regression coefficient from the multivariable analysis [29]. Subsequently, these parameters were used to create a nomogram of the best model which was developed according to the TRIPOD statement [30].

In order to put our findings in perspective with previous studies that did not use competing risk analysis, we also performed univariate analysis using the Kaplan-Meier method with log-rank test and multivariable Cox proportional hazards analysis.

Two-sided p-values <0.05 were considered statistically significant. A c-statistic of <0.50 is equal to chance, a c-statistic of >0.7 is considered a useful test. Analyses were performed using SPSS for Windows version 22 (IBM Corp., Armonk, NY, USA) and the RMS package in R version 3.03 (http://www.r-project.org).

RESULTS

Patients

In total, 841 adult patients with cirrhosis were listed during the study period. A CT examination within 90 days of the listing date was available for 585 of the 841 (69.6%) patients; all others were excluded. Hence, the study cohort comprised 585 patients. The majority was male (n=404, 69.1%) and the median MELD score was 14 (IQR 9-19). A malignancy (i.e. HCC or perihilar cholangiocarcinoma (PHC)) was diagnosed in 193 (33.0%) patients. All baseline characteristics are shown in table 1. Baseline characteristics did not significantly differ between the included and excluded patients, except for median serum sodium (138 mmol/l versus 140 mmol/l, p=0.001), median age (56 [IQR 48-62] versus 54 [IQR 47-60] years, p=0.049), and the distribution of etiology (p=0.024), which was caused by a higher proportion of HCC/PHC among included patients (33.0% versus 20.9%, p=0.001). In total, 425 patients (72.6%) underwent liver transplantation, 15 (2.6%) were still on the waiting list at the end of the study period, and 145 (24.8%) were removed for reasons indicated in table 2. Of the latter group, 90 (15.4%) died on the waiting list or were removed and consequently died within 30 months from removal. The median time between waiting list placement and CT was 31 (IQR 15-51) days. After a median follow-up of 54 months, median overall survival in transplanted patients was 117 months (95% CI 73-160 months). The 3-month, 1-year, 3-year, and 5-year post-transplant overall survival rates were: 95.7%, 90.3%, 83.0%, 79.7%, respectively.

Sarcopenia$_M$ was observed in 254 patients (43.4%) (table 1). Patients with Sarcopenia$_M$ had a significantly lower BMI (p=0.001), a higher MELD score (p<0.001), and a higher MELDNa score (p=0.001). Furthermore, patients with Sarcopenia$_M$ experienced more liver-related complications before waiting list placement compared with patients without sarcopenia (73.2% versus 62.8%, p=0.008). Sarcopenia$_C$ was observed in 266 patients (45.5%). The median MELD-Sarcopenia score was 18 [IQR 12-26].

The association between sarcopenia and waiting list mortality

The median time on the waiting list was 7 months (IQR 3-12) and this was significantly shorter in patients with Sarcopenia$_M$ than patients without Sarcopenia$_M$ (6 [IQR 2-10] versus 8 [IQR 4-14], p<0.001). The proportion of patients who underwent liver transplantation did not differ between patients with and without Sarcopenia$_M$ (71.7% versus 73.4%, p=0.636), whereas the proportion of patients who were removed from the waiting list or died was significantly higher in patients with sarcopenia (20.5% versus 11.5%, p=0.003).

Table 1. Baseline characteristics. Data are shown as median with interquartile ranges (IQR) or counts with percentages. Abbreviations: BMI, Body Mass Index; MELD: Model for End-stage Liver Disease; INR; International Normalized Ratio. * Comparison of patients with Sarcopenia$_M$ vs. without Sarcopenia$_M$.

	All patients N = 585	**Sarcopenia$_M$ N = 254 (43.4%)**	**No Sarcopenia$_M$ N = 331 (56.6%)**	**p-value***
Sex, males	404 (69.1)	169 (66.5)	235 (71.0)	0.247
Age, years	56 (48-62)	57 (48-62)	55 (48-61)	0.171
BMI, kg/m^2	25.7 (22.9-29.3)	25.5 (22.4-28.1)	26.2 (23.4-30.1)	0.001
Primary aetiology of cirrhosis				
Alcoholic	91 (15.6)	48 (18.9)	43 (13.0)	0.148
Hepatitis B virus	16 (2.7)	6 (2.4)	10 (3.0)	
Hepatitis C virus	36 (6.2)	17 (6.7)	19 (5.7)	
PSC/PBC	134 (22.9)	61 (24.0)	73 (22.1)	
HCC/Perihilar cholangiocarcinoma	193 (33.0)	71 (28.0)	122 (36.9)	
NASH	27 (4.6)	16 (6.3)	11 (3.3)	
Cryptogenic	31 (5.3)	13 (5.1)	18 (5.4)	
Auto-immune hepatitis	18 (3.1)	9 (3.5)	9 (2.7)	
Other	39 (6.7)	13 (5.1)	26 (7.9)	
MELD	14 (9-19)	16 (11-20)	13 (9-17)	<0.00
MELDNa	16 (10-22)	18 (11-23)	14 (9-21)	0.001
Bilirubin, µmol/L	45 (20-98)	53 (24-122)	40 (18-87)	0.006
Creatinine, µmol/L	72 (60-92)	74 (59-94)	71 (60-88)	0.235
INR	1.3 (1.1-1.5)	1.3 (1.2-1.7)	1.3 (1.1-1.4)	0.005
Albumin, g/L	34 (30-40)	34 (30-39)	35 (30-41)	0.210
Sodium, mmol/L	134 (138-141)	137 (133-141)	139 (135-141)	0.022
Complications before waiting list placement				
Any	394 (67.4)	186 (73.2)	208 (62.8)	0.008
Ascites	347 (59.9)	169 (67.3)	178 (54.3)	0.001
Spontaneous bacterial peritonitis	94 (16.2)	49 (19.5)	45 (13.6)	0.056
Hepatic encephalopathy	152 (26.3)	84 (33.6)	68 (20.7)	<0.001
Esophageal variceal bleeding	133 (22.9)	64 (25.5)	69 (21.0)	0.199

Using cumulative function incidence analysis, a significant difference in waiting list survival was found between patients with and without Sarcopenia$_M$ (p=0.003; table 3, figure 2), but not between patients with and without Sarcopenia$_C$ (p=0.350; figure 3). The Kaplan-Meier survival curves showed comparable findings (Supplementary figures 1 and 2).

Table 2. Reasons for removal from the waiting list. Other indications included patient preference, substance abuse, no indication after hemihepatectomy, and the development of Alzheimer's disease or other malignancies.

Removal Reason	**n (%)**
Liver transplantation	425 (72.6)
Clinical deterioration / mortality	80 (13.7)
Progression of HCC beyond Milan criteria	42 (7.2)
Improved liver function / stable situation	15 (2.6)
Other	8 (1.4)
Not removed, still on waiting list (31 December 2016)	15 (2.6)

Abbreviations: HCC, Hepatocellular Carcinoma.

Table 3. Performance of the various scores and sarcopenia classifications using competing risk analysis. The presented c-indices show the discriminative value to predict 3-month waiting list mortality.

Score	**sHR**	**95% CI**	**p-value**	**C-index**
MELD	1.09	1.06-1.11	<0.001	0.839
MELDNa	1.05	1.03-1.08	<0.001	0.824
Sarcopenia$_M$	1.88	1.24-2.86	0.003	0.598
Sarcopenia$_C$	1.22	0.81-1.83	0.350	0.515
MELD	1.09	1.06-1.11	<0.001	0.834
Sarcopenia$_M$	1.69	1.09-2.61	0.018	
MELDNa	1.05	1.03-1.08	<0.001	0.798
Sarcopenia$_M$	1.49	0.94-2.36	0.092	
MELD	1.09	1.06-1.11	<0.001	0.835
Sarcopenia$_C$	1.12	0.73-1.72	0.610	
MELDNa	1.06	1.03-1.08	<0.001	0.792
Sarcopenia$_C$	0.93	0.59-1.47	0.750	
MELD-Sarcopenia [1]	1.08	1.05-1.10	<0.001	0.820

Abbreviations: sHR, subdistribution Hazard Ratio; CI, Confidence Interval; MELD, Model for End-stage Liver Disease; C-index, Concordance index; SE, Standard Error.

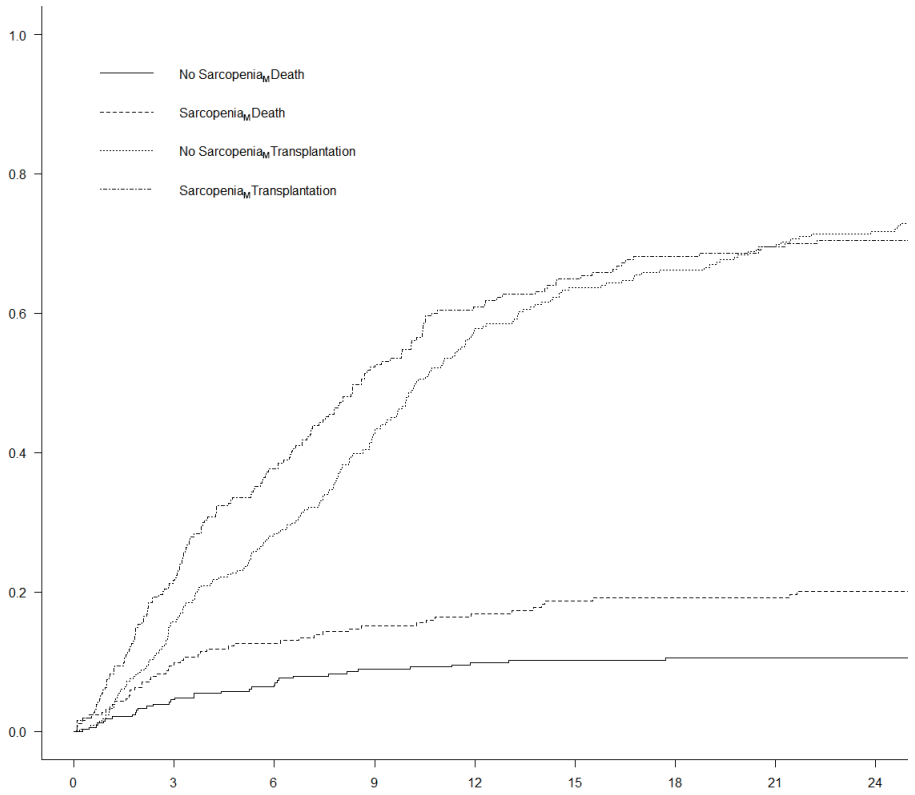

Figure 2. Cumulative incidence functions for patients with and without Sarcopenia$_M$. The median waiting list survival was significantly shorter in patients with sarcopenia (HR 1.88 [95% CI 1.24-2.86], p=0.003).

Mortality was significantly higher in patients with Sarcopenia$_M$ than in patients without Sarcopenia$_M$ after 1 month (35.0% versus 20.8%, p<0.001), 3 months (48.4% versus 32.3%, p<0.001), 1 year (84.3% versus 75.5%, p=0.010), and 3 years (96.9% versus 93.7%, p=0.078), whereas no differences were observed for patients with and without Sarcopenia$_C$.

Performance of the various scores

table 3 shows the performance of the various scores, with the corresponding c-indices for 3-month waiting list mortality. Sarcopenia$_M$ adjusted for the MELD score showed an excellent discriminative performance to predict 3-month mortality (c-index 0.834). However, it did not exceed the discriminative performance of the MELD score alone

(c-index 0.839). The discriminative performance of Sarcopenia$_C$ was poor (c-index 0.515), but increased to 0.835 when adjusted for MELD score. The c-index for the MELD-Sarcopenia score was 0.820. Supplementary table 1 shows comparable data based on Cox regression analysis. An overestimation of the discriminative value was observed compared with competing risk analysis.

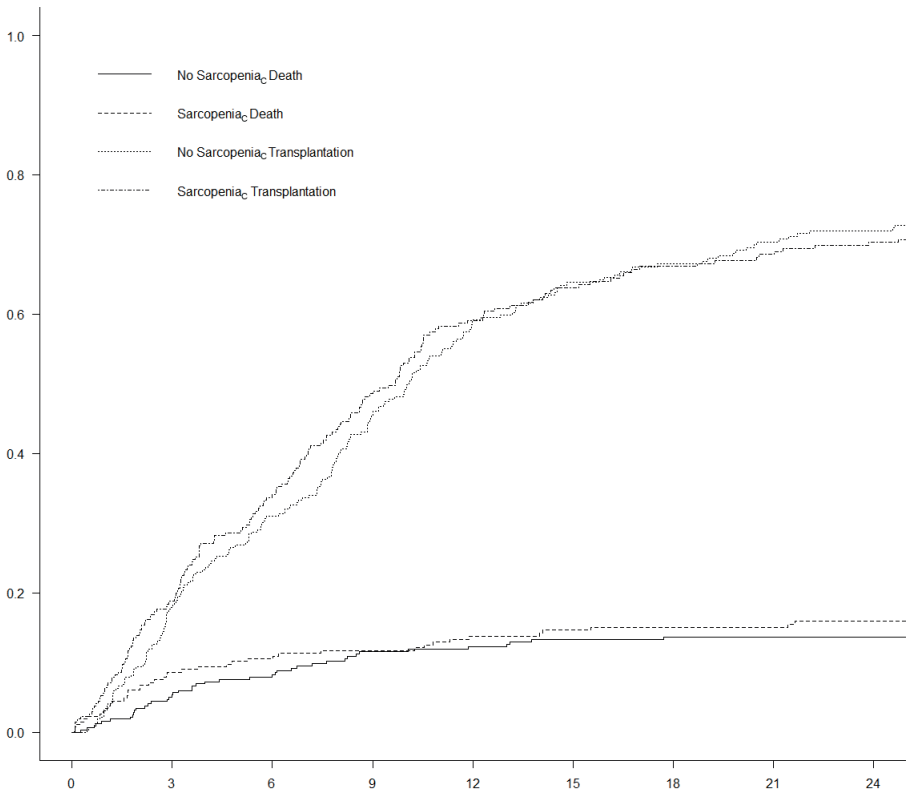

Figure 3. Cumulative incidence functions for patients with and without Sarcopenia$_C$. No survival differences were found between patients with and without sarcopenia (HR 1.22 [95% CI 0.81-1.83, p=0.350]).

A multivariable competing risk analysis and the development of a nomogram

After backward selection using the AIC, the model with the highest discriminative performance to predict waiting list mortality included MELD score (sHR 1.09 [95% CI 1.06-1.12], p<0.001), hepatic encephalopathy before listing (sHR 1.80 [95% CI 1.12-2.87], p=0.014), age (sHR 1.02 [95% CI 0.99-1.05], p=0.085) and $Sarcopenia_M$ (sHR 1.51 [95% CI 0.97-2.34], p=0.067), with a c-index of 0.851 (table 4). The nomogram of this model, is depicted in figure 4.

We performed stratified analyses based on low and high MELD-scores (cut-off MELD score 15). The independent additive effect of sarcopenia was strongly present in the group with low MELD-scores (n=342, sHR 2.10 [95% CI 1.05-4.2], p=0.035) but not in the group with high MELD-scores (n=243, sHR 1.30 [95% CI 0.75-2.26], p=0.349). The calibration of the nomogram was assessed and showed fair calibration, particularly for patients at low risk for waiting list mortality (figures 5a and 5b).

Table 4. Multivariable competing risk analysis according to Fine and Gray for waiting list survival. Variables were selected using a backwards stepwise selection based on the Akaika Information Criterion (AIC). The model was based on 578 patients due to missing data on the occurrence of hepatic encephalopathy before listing in 7 patients. The discriminative value of the model was excellent with a c-index of 0.851 for 3-month waiting list mortality.

	sHR	95% CI	p-value
MELD	1.09	1.06-1.12	<0.001
$Sarcopenia_M$	1.51	0.97-2.34	0.067
Age, years	1.80	1.12-2.87	0.014
Hepatic encephalopathy before listing	1.02	1.00-1.05	0.085

Abbreviations: sHR, subdistribution Hazard Ratio; CI, Confidence Interval; MELD, Model for End-stage Liver Disease.

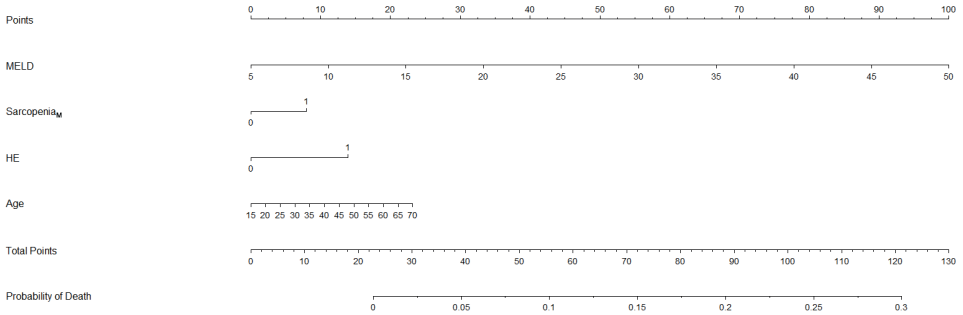

Figure 4. Nomogram to predict waiting list survival. The sum of the specific weighted scores was associated with decreased survival.

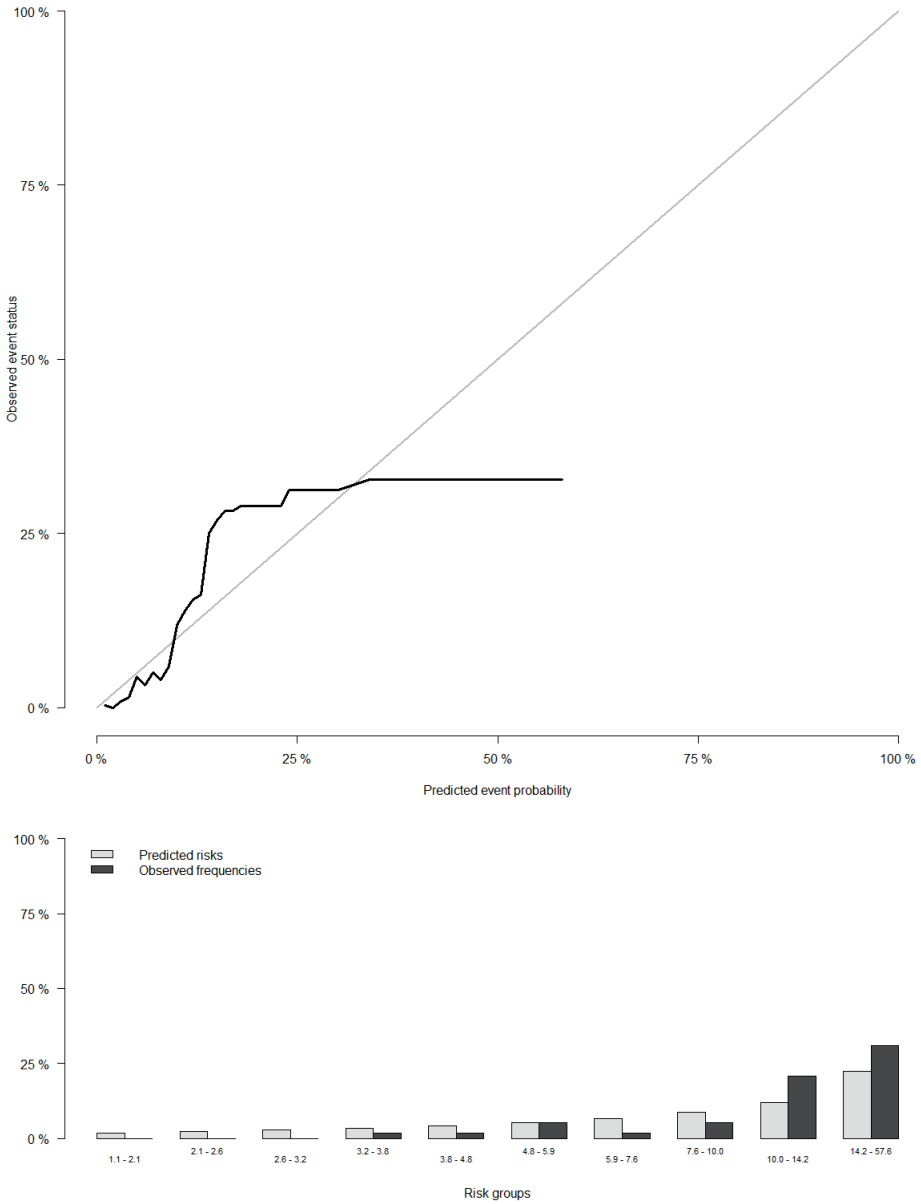

Figure 5a. Calibration plot (A) and calibration bar plot (B) for the nomogram. Particularly patients with a low risk on waiting list mortality are accurately predicted using the nomogram.

DISCUSSION

In this study we found that sarcopenia was a risk factor for waiting list mortality in a cohort of Western-European liver transplant candidates with cirrhosis. The MELD-Sarcopenia score had a discriminative performance (c-index 0.82 for 3-month mortality), which was comparable with the discriminative performance in the training cohort of the original study (c-index 0.85) [10], but lower than the original MELD score (c-index 0.84) in our cohort. A final competing risk model included Sarcopenia$_M$, MELD score, age, and presence of hepatic encephalopathy before listing as predictors for waiting list mortality. This may be attributed to the fact that prioritization and allocation of donor organs and transplant candidates as well as waiting list mortality are all strongly related to the MELD score, resulting in a self-fulfilling prophecy.

The association between sarcopenia and mortality in patients evaluated or listed for liver transplantation has been previously described [6, 11-15, 25]. However, most studies have been performed in predominantly (and partly overlapping) North-American populations [11-15,25]. Furthermore, most studies included a relatively small number of patients varying from n=59 to n=213 [11-14, 16], and moreover none of the studies took competing risks on the waiting list into account. Which is important while we show that the prognostic value of the results obtained by 'normal' cox-proportional hazard analyses as opposed to competing risk analyses, overestimated rather than underestimated survival. The current study is the first multicenter Western-European study with a large number of patients. In our cohort median waiting list period was 7 (IQR 3-12) months, after which 72.6% of patients underwent transplantation and 15.4% were removed from or died on the waiting list or within 3 months after removal. In the cohort of Carey *et al.*, including 396 listed patients, only 50% underwent transplantation and 28% were delisted or died [25]. Similarly, in the cohort of Montano-Loza *et al.*, which included cirrhotic patients with a comparable MELD score, only 34% underwent transplantation and 39% died with a median follow-up of 11 months [10]. The difference in outcomes as compared to our study could be explained by the fact that the cohort of Montano-Loza consists of cirrhotic patients who were evaluated for liver transplantation, but not necessarily listed yet. The greater homogeneity of our cohort may also explain some of the differences, amongst others the higher discriminative performance of the MELD-Sarcopenia score using both Cox regression and competing risk analysis.

Apart from the MELD-Sarcopenia score, others proposed a score for waiting list mortality with muscle mass included in the MELD score: the MELD-psoas score. In this score, only the psoas muscle area was measured rather than the cross-sectional muscle area [6]. The cross-sectional measurements have previously been validated whereas results regarding

accuracy of psoas muscle measurements are conflicting. Two? recent studies suggested that psoas muscle measurements are inferior to total skeletal muscle measurements [31,32]. But, in contrast, the psoas muscle is successfully used in liver transplantation patients according to a French study [33]. In addition, a Japanese study also included measures of skeletal muscle in the MELD score (Muscle-MELD score) to predict mortality after living-donor liver transplantation (LDLT) [34]. However, today, it remains unknown which of the two measurements most accurately reflects total body skeletal muscle mass, and subsequently predicts mortality.

To date, there are no generally accepted cut-off values to classify patients with sarcopenia. The most frequently used definition is that of Martin *et al.* (Sarcopenia$_M$ [24]), who created cut-off values for sarcopenia to predict survival in cancer patients. These are based on sex and BMI (for men). Validity of the use of BMI is argued; as a high proportion of liver transplant candidates suffers from ascites (59.9% in our cohort), subsequently BMI will be overestimated and hence sarcopenia could be overestimated in male patients. Nevertheless, the discriminative performance using these cut-off values was excellent in our cohort, whereas no survival differences were found with the cut-off values of Carey *et al.* (Sarcopenia$_C$ [25]). This is of interest, while the cut-off values in the latter study were defined to predict waiting list mortality in liver transplant candidates, particularly. After adjusting sarcopenia$_c$ [25] for the MELD score, the discriminative performance drastically increased, which underlines the strong predictive power of the MELD score.

As reflected by a higher proportion of liver-related complications, sarcopenia is strongly correlated with severity of liver disease. It may therefore contribute to better prioritising liver transplant candidates, as these patients are at risk to die or to deteriorate prematurely and this risk is currently not reflected in their MELD-score. In line with previous results [6], we found that sarcopenia is a stronger prognostic factor in patients with a low compared with patients with a high MELD score (based on a cut-off for MELD score of 15). Therefore, patients with sarcopenia and a low MELD score may be under prioritised in the current allocation system.

Possible explanations as to why liver disease leads to skeletal muscle depletion and sarcopenia are altered food intake, hypermetabolism from chronic disease, altered amino acid profiles, endotoxemia, accelerated starvation and decreased mobility [35]. Moreover, hyperammonia was recently described as an additional mediator in the liver-muscle axis [35]. Skeletal muscle mass serves as storage for ammonia and depletion of muscle mass could therefore explain the increased rate of hepatic encephalopathy in patients with sarcopenia.

Both functional impairment at the moment of listing [8] and a significant functional decline over time during the waiting period (both measured with the Short Physical Performance Battery) have been previously associated with an increased risk of death, independent of the severity of liver disease [7]. Therefore, the waiting list period, or even the liver transplant evaluation period, offer a window of opportunity to improve functional status. Suggested regimens may consist of the use of proteins with low ammoniagenic potential, leucine enriched amino acid supplementation, long-term ammonia lowering strategies and a combination of resistance and endurance exercise to increase muscle mass and function [35].

This study has some limitations we would like to address. Although we used the prospective Eurotransplant registry data with all consecutive patients who have been listed for liver transplantation, some data was collected retrospectively (e.g. serum albumin and sodium levels) which led to missing data in a few cases. However, we tried to overcome bias due to missing data using multiple imputations for these laboratory values. Moreover, CT examinations performed within 90 days from listing were not available in all patients, as the time interval between screening and the eventual listing data greatly varied. Although we internally validated our findings in a prospective, national cohort, external validation may be warranted. Finally, we only have data on CT examinations at the moment of listing instead of follow-up data on skeletal muscle wasting along the waiting list period. One of the major advantages of the MELD score is the ability to easily calculate the MELD score at bedside. Nevertheless, evaluation of skeletal muscle mass will only take a couple of extra minutes extra per patient but could be of additional value for patients' quality of life and survival [36]. For this purpose, automated software is currently under development [36, 37].

In conclusion, sarcopenia is strongly associated with waiting list survival in liver transplant candidates with cirrhosis. Our model may be used to identify patients at risk for waiting list mortality, particularly those with otherwise lower MELD scores. These patients may benefit from more intensive follow-up, monitoring, and training programs or prioritising on the waiting list.

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REFERENCES

1. Freeman RB, Jr., Wiesner RH, Harper A, McDiarmid SV, Lake J, Edwards E, et al. The new liver allocation system: moving toward evidence-based transplantation policy. Liver Transpl 2002; 8: 851-8.
2. Kamath PS, Kim WR, Advanced Liver Disease Study G. The model for end-stage liver disease (MELD). Hepatology 2007; 45: 797-805.
3. Kim WR, Biggins SW, Kremers WK, Wiesner RH, Kamath PS, Benson JT, et al. Hyponatremia and mortality among patients on the liver-transplant waiting list. The New England journal of medicine 2008; 359: 1018-26.
4. Myers RP, Shaheen AA, Faris P, Aspinall AI, Burak KW. Revision of MELD to include serum albumin improves prediction of mortality on the liver transplant waiting list. PLoS One 2013; 8: e51926.
5. Myers RP, Tandon P, Ney M, Meeberg G, Faris P, Shaheen AA, et al. Validation of the five-variable Model for End-stage Liver Disease (5vMELD) for prediction of mortality on the liver transplant waiting list. Liver Int 2014; 34: 1176-83.
6. Durand F, Buyse S, Francoz C, Laouenan C, Bruno O, Belghiti J, et al. Prognostic value of muscle atrophy in cirrhosis using psoas muscle thickness on computed tomography. J Hepatol 2014; 60: 1151-7.
7. Lai JC, Dodge JL, Sen S, Covinsky K, Feng S. Functional decline in patients with cirrhosis awaiting liver transplantation: Results from the functional assessment in liver transplantation (FrAILT) study. Hepatology 2016; 63: 574-80.
8. Lai JC, Feng S, Terrault NA, Lizaola B, Hayssen H, Covinsky K. Frailty predicts waitlist mortality in liver transplant candidates. Am J Transplant 2014; 14: 1870-9.
9. van Vugt JL, Levolger S, de Bruin RW, van Rosmalen J, Metselaar HJ, IJzermans JN. Systematic review and meta-analysis of the impact of computed tomography assessed skeletal muscle mass on outcome in patients awaiting or undergoing liver transplantation. Am J Transplant 2016; 16: 2277-92.
10. Montano-Loza AJ, Duarte-Rojo A, Meza-Junco J, Baracos VE, Sawyer MB, Pang JX, et al. Inclusion of Sarcopenia Within MELD (MELD-Sarcopenia) and the Prediction of Mortality in Patients With Cirrhosis. Clinical and translational gastroenterology 2015; 6: e102.
11. Meza-Junco J, Montano-Loza AJ, Baracos VE, Prado CM, Bain VG, Beaumont C, et al. Sarcopenia as a prognostic index of nutritional status in concurrent cirrhosis and hepatocellular carcinoma. J Clin Gastroenterol 2013; 47: 861-70.
12. Montano-Loza AJ, Meza-Junco J, Prado CM, Lieffers JR, Baracos VE, Bain VG, et al. Muscle wasting is associated with mortality in patients with cirrhosis. Clin Gastroenterol Hepatol 2012; 10: 166-73, 73 e1.

13. Tandon P, Ney M, Irwin I, Ma MM, Gramlich L, Bain VG, et al. Severe muscle depletion in patients on the liver transplant wait list: its prevalence and independent prognostic value. Liver Transpl 2012; 18: 1209-16.
14. Yadav A, Chang YH, Carpenter S, Silva AC, Rakela J, Aqel BA, et al. Relationship between sarcopenia, six-minute walk distance and health-related quality of life in liver transplant candidates. Clin Transplant 2015; 29: 134-41.
15. Montano-Loza AJ, Angulo P, Meza-Junco J, Prado CM, Sawyer MB, Beaumont C, et al. Sarcopenic obesity and myosteatosis are associated with higher mortality in patients with cirrhosis. J Cachexia Sarcopenia Muscle 2016; 7: 126-35.
16. Giusto M, Lattanzi B, Albanese C, Galtieri A, Farcomeni A, Giannelli V, et al. Sarcopenia in liver cirrhosis: the role of computed tomography scan for the assessment of muscle mass compared with dual-energy X-ray absorptiometry and anthropometry. Eur J Gastroenterol Hepatol 2015; 27: 328-34.
17. Noordzij M, Leffondre K, van Stralen KJ, Zoccali C, Dekker FW, Jager KJ. When do we need competing risks methods for survival analysis in nephrology? Nephrol Dial Transplant 2013; 28: 2670-7.
18. Sapir-Pichhadze R, Pintilie M, Tinckam KJ, Laupacis A, Logan AG, Beyene J, et al. Survival Analysis in the Presence of Competing Risks: The Example of Waitlisted Kidney Transplant Candidates. Am J Transplant 2016; 16: 1958-66.
19. Eurotransplant. 2017 [27-04-2017]; Available from: www.eurotransplant.org.
20. Mazzaferro V, Regalia E, Doci R, Andreola S, Pulvirenti A, Bozzetti F, et al. Liver transplantation for the treatment of small hepatocellular carcinomas in patients with cirrhosis. The New England journal of medicine 1996; 334: 693-9.
21. van Vledder MG, Levolger S, Ayez N, Verhoef C, Tran TC, IJzermans JN. Body composition and outcome in patients undergoing resection of colorectal liver metastases. Br J Surg 2012; 99: 550-7.
22. Shen W, Punyanitya M, Wang Z, Gallagher D, St-Onge MP, Albu J, et al. Total body skeletal muscle and adipose tissue volumes: estimation from a single abdominal cross-sectional image. J Appl Physiol (1985) 2004; 97: 2333-8.
23. van Vugt JL, Levolger S, Gharbharan A, Koek M, Niessen WJ, Burger JW, et al. A comparative study of software programmes for cross-sectional skeletal muscle and adipose tissue measurements on abdominal computed tomography scans of rectal cancer patients. J Cachexia Sarcopenia Muscle 2017; 8: 285-97.
24. Martin L, Birdsell L, Macdonald N, Reiman T, Clandinin MT, McCargar LJ, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol 2013; 31: 1539-47.
25. Carey EJ, Lai JC, Wang CW, Dasarathy S, Lobach I, Montano-Loza AJ, et al. A multi-center study to define sarcopenia in patients with end-stage liver disease. Liver Transpl 2017.

26. Wolbers M, Blanche P, Koller MT, Witteman JC, Gerds TA. Concordance for prognostic models with competing risks. Biostatistics 2014; 15: 526-39.

27. Wolbers M, Koller MT, Witteman JC, Steyerberg EW. Prognostic models with competing risks: methods and application to coronary risk prediction. Epidemiology 2009; 20: 555-61.

28. Fine J, Gray R. A proportional hazards model for the subdistribution of a competing risk. J Am Stat Assoc 1999; 94: 496-509.

29. Buettner S, Wagner D, Kim Y, Margonis GA, Makary MA, Wilson A, et al. Inclusion of Sarcopenia Outperforms the Modified Frailty Index in Predicting 1-Year Mortality among 1,326 Patients Undergoing Gastrointestinal Surgery for a Malignant Indication. Journal of the American College of Surgeons 2016; 222: 397-407e2.

30. Collins GS, Reitsma JB, Altman DG, Moons KG. Transparent Reporting of a multivariable prediction model for Individual Prognosis Or Diagnosis (TRIPOD): the TRIPOD Statement. Br J Surg 2015; 102: 148-58.

31. Rutten IJ, Ubachs J, Kruitwagen RFPM, Beets-Tan RGH, Olde Damink SWM, van Gorp T. Abstracts of the 9th International Conference on Cachexia, Sarcopenia, and Muscle Wasting, Berlin, Germany, 10–11 December 2016 (part 2) (1-45: Psoas muscle measurements are inferior to total skeletal muscle measurements in the assessment of sarcopenia in ovarian cancer). J Cachexia Sarcopenia Muscle 2017; 8: 161-83.

32. Baracos VE. Psoas as a sentinel muscle for sarcopenia: a flawed premise. J Cachexia Sarcopenia Muscle 2017; 8: 527-8.

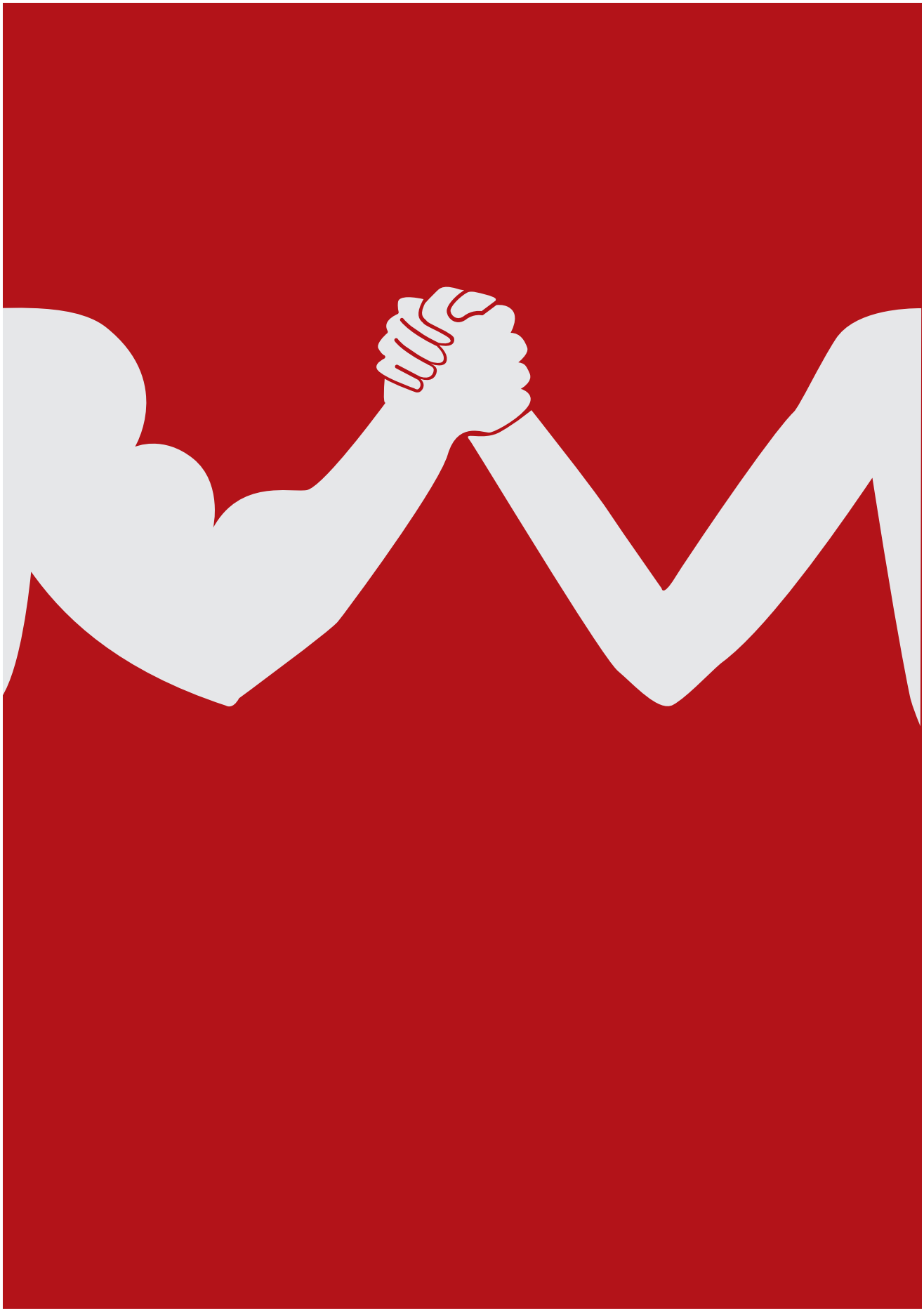
33. Golse N, Bucur PO, Ciacio O, Pittau G, Sa Cunha A, Adam R, et al. A new definition of sarcopenia in patients with cirrhosis undergoing liver transplantation. Liver Transpl 2017; 23: 143-54.

34. Hamaguchi Y, Kaido T, Okumura S, Kobayashi A, Shirai H, Yagi S, et al. Proposal of Muscle-MELD Score, Including Muscularity, for Prediction of Mortality After Living Donor Liver Transplantation. Transplantation 2016; 100: 2416-23.

35. Dasarathy S, Merli M. Sarcopenia from mechanism to diagnosis and treatment in liver disease. J Hepatol 2016; 65: 1232-44.

36. Takahashi N, Sugimoto M, Psutka SP, Chen B, Moynagh MR, Carter RE. Validation study of a new semi-automated software program for CT body composition analysis. Abdom Radiol (NY) 2017.

37. Popuri K, Cobzas D, Esfandiari N, Baracos V, Jagersand M. Body Composition Assessment in Axial CT Images Using FEM-Based Automatic Segmentation of Skeletal Muscle. IEEE Trans Med Imaging 2016; 35: 512-20.



PART V

THE SOCIO-ECONOMIC CONSEQUENCES OF LOW SKELETAL MUSCLE MASS

CHAPTER 12

Low Skeletal Muscle Mass is Associated with Increased Hospital Expenditure in Patients Undergoing Cancer Surgery of the Alimentary Tract

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ABSTRACT

Background: Low skeletal muscle mass is associated with poor postoperative outcomes in cancer patients. Furthermore, it is associated with increased healthcare costs in the United States. We investigated its effect on hospital expenditure in a Western-European healthcare system, with universal access.

Methods: Skeletal muscle mass (assessed on CT) and costs were obtained for patients who underwent curative-intent abdominal cancer surgery. Low skeletal muscle mass was defined based on pre-established cut-offs. The relationship between low skeletal muscle mass and hospital costs was assessed using linear regression analysis and Mann-Whitney U-tests.

Results: 452 patients were included (median age 65, 61.5% males). Patients underwent surgery for colorectal cancer (38.9%), colorectal liver metastases (27.4%), primary liver tumours (23.2%), and pancreatic/periampullary cancer (10.4%). In total, 45.6% had sarcopenia. Median costs were €2,183 higher in patients with low compared with patients with high skeletal muscle mass (€17,144 versus €14,961; p<0.001). Hospital costs incrementally increased with lower sex-specific skeletal muscle mass quartiles (p=0.029). After adjustment for confounders, low skeletal muscle mass was associated with a cost increase of €4,061 (p=0.015).

Conclusions: Low skeletal muscle mass was independently associated with increased hospital costs of about €4,000 per patient. Strategies to reduce skeletal muscle wasting could reduce hospital costs in an era of incremental healthcare costs and an increasingly aging population.

INTRODUCTION

Low skeletal muscle mass is a strong predictor of complications, reduced therapy effect, impaired survival in gastrointestinal and hepatopancreatobiliary cancer patients undergoing surgery [1-3], and dose-limiting chemotherapy toxicity [4-6]. In addition to clinical outcome, recent studies from the United States showed that low skeletal muscle mass is associated with increased healthcare costs [7-9].

However, the lower accessibility and higher uninsured rate of the American healthcare system, which is known to be greatly affected by income [10], may impair extrapolation to other healthcare systems with universal access [11-13]. Because European healthcare costs are increasingly rising, we aimed to assess the effect of low skeletal muscle mass on hospital costs of patients undergoing curative-intent surgery for abdominal cancer in a Western-European healthcare system.

MATERIALS AND METHODS

Patients and data acquisition

In this retrospective study, patients aged 18 years or older who underwent curative surgery for gastrointestinal or hepatopancreatobiliary cancers in our center were identified from various databases (including colorectal carcinoma [14-16], hepatocellular carcinoma [17], colorectal liver metastases [18], perihilar [19] or intrahepatic [19] cholangiocarcinoma, and pancreatic or periampullary cancer [20]). Patients underwent surgery between 2005 and 2015. Only patients with a CT imaging within 90 days preoperatively were included. Demographics and patient characteristics were collected from electronic patient files. Preoperative physical status was assessed using the American Society of Anesthesiologists (ASA) score [21]. Overweight was defined as a body mass index (BMI) ≥ 25 kg/m^2, according to the definition of the World Health Organization [22]. Postoperative complications were identified in the medical patient files and recorded. Severity of postoperative complications was scored according to the Clavien-Dindo classification [23]. Severe complications were defined as complications with grade 3a or higher. Postoperative mortality was defined as mortality during hospital stay or within 30 days after surgery. Length of hospital stay was calculated by counting admission days from the day of surgery. An hospital stay exceeding 7 days was considered a prolonged hospital stay. Patients were divided in two groups according to the extent of surgery: 1. Major surgery (i.e. the resection of at least two hepatic segments and a wedge resection or the resection of at least three hepatic segments, pancreatic surgery, and pelvic exenteration for locally advanced rectal cancer); 2. Minor surgery (i.e. one- or two hepatic segment resections, and colorectal resections). The study was approved by the Medical Ethical Committee of Erasmus MC University Medical Center and a waiver for informed consent was granted.

Skeletal muscle mass measurements

Skeletal muscle mass was measured on computed tomography (CT) examinations that were routinely performed as part of diagnostic or preoperative work-up, as previously described [18]. In short, the cross-sectional skeletal muscle area was measured at the level of the third lumbar vertebra on a slice on which both transversal processes were visible using a Hounsfield unit threshold of -30 to +150. The cross-sectional muscle area was then corrected for patients' height squared, as is conventional for body composition measurements, resulting in the skeletal muscle index (SMI, cm^2/m^2). Patients were classified as having low skeletal muscle mass according to predefined cut-off values established by Martin and colleagues: males with body mass index (BMI) <25: <43 cm^2/m^2, males with BMI ≥25: <53 cm^2/m^2, females: <41 cm^2/m^2 [24]. Furthermore, patients were

divided in sex-specific quartiles based on their skeletal muscle index. In patients who underwent induction or neoadjuvant chemo(radio)therapy, the CT after chemotherapy was used.

Cost analyses

Costs were abstracted from the hospital's electronic accounting system, which is based on activity based costing. All costs that were made during the index admission for surgery, including surgical costs and all costs in the postoperative period and independent of (medical) discipline, were included. Total costs were calculated by the sum of all unit cost-prices. These cost prices are covering the full cost prices. Financial data was limited to expenses within the index admission and did not include costs after discharge from the index admission or costs made outside our center. Adjustment for inflation was performed by indexing all cost-prices to the year 2015 according to data of the Dutch Healthcare Authority. All financial data are reported in Euros (€).

Statistical analyses

Categorical data are reported as counts with percentages, whereas continuous data are reported as median with interquartile range (IQR) or mean with standard deviation (SD), depending on their normality of distribution. Categorical data was compared using the chi-squared test. The Mann-Whitney-U and Kruskall-Wallis H tests was used to compare hospital costs between groups. A multivariable linear regression analysis was performed to investigate the independent association of the presence of low skeletal muscle mass with increased costs after correction for possible confounding and clinically relevant factors. Sex-specific skeletal muscle mass quartiles were compared using one-way ANOVA analysis. Continuous baseline and outcome characteristics were dichotomized, according to clinically relevant cut-off points, and subgroup analyses were performed to investigate the effect of low skeletal muscle mass on total hospital costs in various subgroups. Patients who died during hospital admission or within 30 days postoperative were excluded from the subgroup analyses based on length of hospital stay. Two-sided p-values <0.05 were considered statistically significant. All analyses were performed using SPSS for Windows (IBM Corp., Armonk, NY, USA), version 22.

RESULTS

Patients

In total, 602 patients were identified. Of these patients, no preoperative CT was available in 83 patients (13.8%) and no costs were available for 18 (3.0%) patients. In 49 patients (8.1%) the CT was not performed within 90 days preoperatively. This resulted in 452 patients (75.1%) who formed the study cohort, of whom 278 (61.5%) were male and 174 (38.5%) female with a median age of 65 (IQR 58-71) years. Most patients underwent surgery for colorectal carcinoma (n=176, 38.9%), followed by colorectal liver metastases (n=124, 27.4%), hepatocellular carcinoma (n=53, 11.7%), pancreatic or periampullary cancer (n=47, 10.4%), intrahepatic cholangiocarcinoma (n=32, 7.1%), and perihilar cholangiocarcinoma (n=20, 4.4%). Baseline characteristics are summarized in table 1.

The association between low skeletal muscle mass and treatment outcomes

Almost half of our cohort (45.6%, n=206) had low skeletal muscle mass. Patients with low skeletal muscle mass experienced more postoperative complications compared with patients with normal skeletal muscle mass (55.1% versus 44.9%, p=0.031), and showed an increased length of hospital stay (median 9 [IQR 7-14] versus 8 [IQR 6-12] days, p=0.005). Non-significant differences were observed for severe postoperative complications and postoperative mortality (table 2).

Total hospital costs in patients with low and normal skeletal muscle mass

The median total hospital costs per patient were €16,021 (IQR 11,714-23,381). Total hospital costs significantly differed between cancer types, and were highest in patients operated on for perihilar cholangiocarcinoma and lowest in patients who underwent surgery for colorectal liver metastases (table 3). Patients undergoing major surgery had significantly higher costs compared with patients undergoing minor surgery (€21,124 [IQR 14,618-29,884] versus €12,989 [IQR 10,029-16,736], p<0.001).

Total costs were higher for patients with low skeletal muscle mass compared with patients with normal skeletal muscle mass (€17,144 [IQR 12,694-25,102] versus €14,961 [IQR 10,744-21,200]; p<0.001), resulting in a median difference of €2,183 (12.7%). In univariable linear regression analysis, presence of low skeletal muscle mass was associated with a cost increase of €4,979 (p=0.002). After classifying patients in sex-specific quartiles based on skeletal muscle mass, a decreasing trend in median total hospital costs per patient was observed per incremental quartile (1st quartile €18,320; 2nd quartile €16,172; 3rd quartile €15,501; 4th quartile €14,655; p=0.029, figure 1).

Table 1. Baseline characteristics.

	All patients n=452	Low skeletal muscle mass n=206	Normal skeletal muscle mass n=246	p-value
Sex				
Males	278 (61.5)	111 (53.9)	167 (67.9)	0.002
Females	174 (38.5)	95 (46.1)	79 (32.1)	
Age (years)	64.7 (57.8-71.4)	65.1 (58.3-73.0)	64.5 (57.2-70.3)	0.069
BMI (kg/m^2)*	25.2 (22.7-27.9)	25.1 (21.8-27.2)	25.2 (23.4-28.6)	0.008
ASA classification#				
1-2	334 (79.3)	154 (78.6)	180 (80.0)	0.718
3-4	87 (20.7)	42 (21.4)	45 (20.0)	
Cancer diagnosis				
Colorectal	176 (38.9)	75 (36.4)	101 (41.1)	<0.001
CRLM	124 (27.4)	38 (18.4)	86 (35.0)	
HCC	53 (11.7)	27 (13.1)	26 (10.6)	
Pancreatic/periampullary	47 (10.4)	34 (16.5)	13 (5.3)	
ICC	32 (7.1)	19 (9.2)	13 (5.3)	
PHC	20 (4.4)	13 (6.3)	7 (2.8)	

Abbreviations: BMI, Body Mass Index (* missing for 2 patients); ASA, American Society for Anesthesiologists (# missing for 34 patients); CRLM, Colorectal Liver Metastases; HCC, Hepatocellular Carcinoma; ICC, Intrahepatic Cholangiocarcinoma; PHC, Perihilar Cholangiocarcinoma.

Table 2. Treatment outcomes.

	Low skeletal muscle mass n=206	Normal skeletal muscle mass n=246	p-value
Any postoperative complication	113 (55.1)	110 (44.9)	0.031
Any severe postoperative complication*	48 (23.4)	47 (19.2)	0.273
Postoperative mortality#	15 (7.3)	9 (3.7)	0.087
Length of hospital stay (days)‡	9 (7-14)	8 (6-12)	0.005

* Defined as Clavien-Dindo classification ≥ 3a
# Defined as in-hospital or 30-day mortality
‡ Without patients who died in-hospital or within 30 days postoperatively

Table 3. Total hospital costs per cancer type.

Cancer type	Total hospital costs, € (IQR)	p-value
Colorectal	15,121 (11,718-19,945)	<0.001
CRLM	12,431 (8,721-14,679)	
HCC	22,396 (16,368-32,474)	
Pancreatic/periampullary	22,057 (18,509-25,445)	
ICC	30,130 (18,710-40,827)	
PHC	36,542 (28,122-53,126)	

Abbreviations: IQR, Interquartile Range; CRLM, Colorectal Liver Metastases, HCC, Hepatocellular Carcinoma; ICC, Intrahepatic Cholangiocarcinoma; PHC, Perihilar Cholangiocarcinoma.

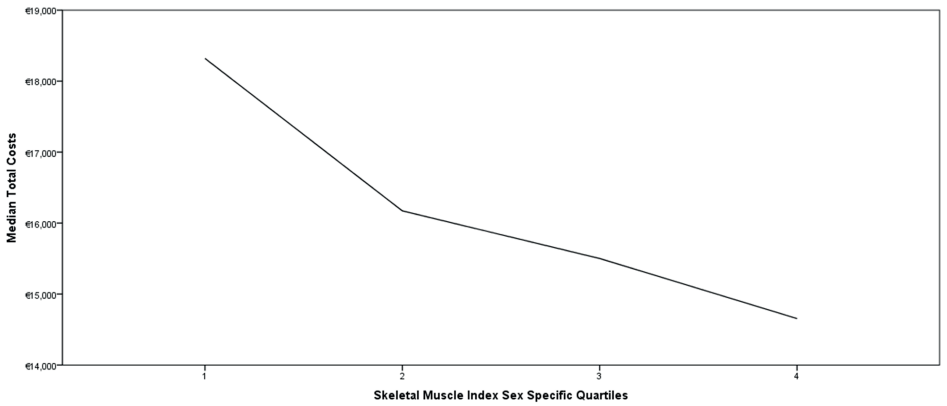

Figure 1. Total hospital costs by skeletal muscle mass in sex-specific quartiles. The total hospital costs significantly decreased per skeletal muscle index sex-specific quartile (p=0.003).

Median hospital costs were also higher for patients with low skeletal muscle mass compared to patients without low skeletal muscle mass within the subgroup of males (€17,823 [IQR 12,816-24,973] versus €15,444 [IQR 10,363-22,396], p=0.006) and females (€16,755 [IQR 12,081-25,257] versus €14,606 [IQR 11,152-20,898], p=0.121) (figure 2a), patients <65 years (€17,184 [IQR 12,619-25,073] versus €14,572 [IQR 10,151-21,250], p=0.025) and ≥65 years (€18,256 [IQR 12,808-25,131] versus €15,490 [IQR 11,060-21,098], p=0.041) (figure 2b) and with overweight (€17,392 [IQR 12,808-23,851] versus €14,667 [IQR 10,277-21,263], p=0.018) and under- or normal weight (€17,410 [IQR 12,457-25,351] versus €15,444 [IQR 10,905-21,179], p=0.048) (figure 2c).

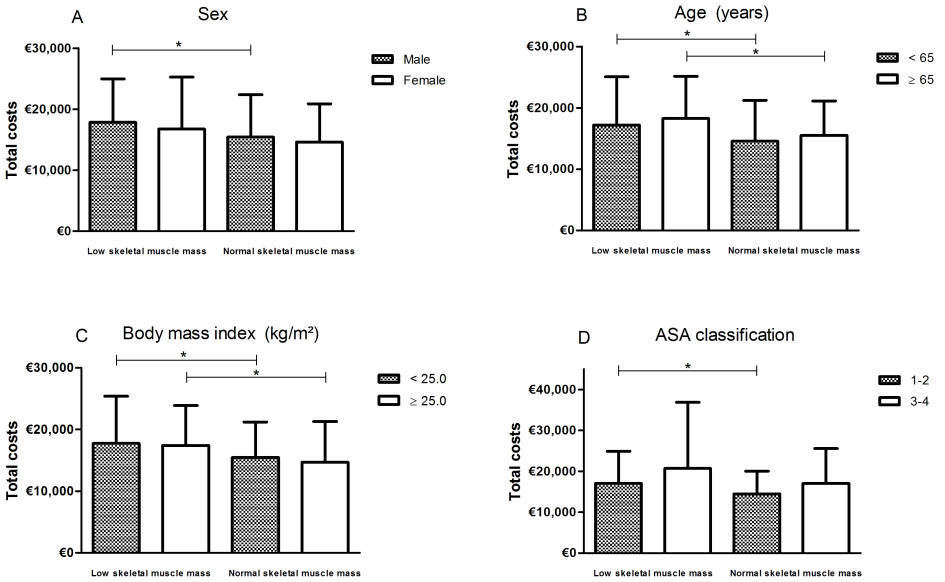

Figure 2. Total hospital costs stratified by the presence of low skeletal muscle mass per patient characteristic. * p<0.05, ** p<0.001

The influence of low skeletal muscle mass and postoperative complications on total hospital costs

Within the group of patients who did not experience any postoperative complication, the median hospitals costs for patients with low skeletal muscle mass were higher compared with the median costs for patients without low skeletal muscle mass (€13,141 [IQR 9,933-18,046] versus €11,715 [IQR 9,161-16,027], p=0.055), figure 3a. The same association was observed in patients who did not experience any severe postoperative complications (€15,448 [IQR 12,079-21,789] in patients with low skeletal muscle mass versus €13,658 [IQR 9,938-18,270] in patients without low skeletal muscle mass, p=0.006), figure 3b. The same effect was observed in patients who did (p=0.084) or did not (p=0.019) die in-hospital or within 30 days postoperatively, and in patients with normal (p=0.041) and prolonged (p=0.434) hospital stay (figure 3d) (figure 3c). Low skeletal muscle mass was associated with increased total hospital costs in the subgroup of patients undergoing major resections (p=0.099), but not within the subgroups of minor resections (p=0.238), figure 4.

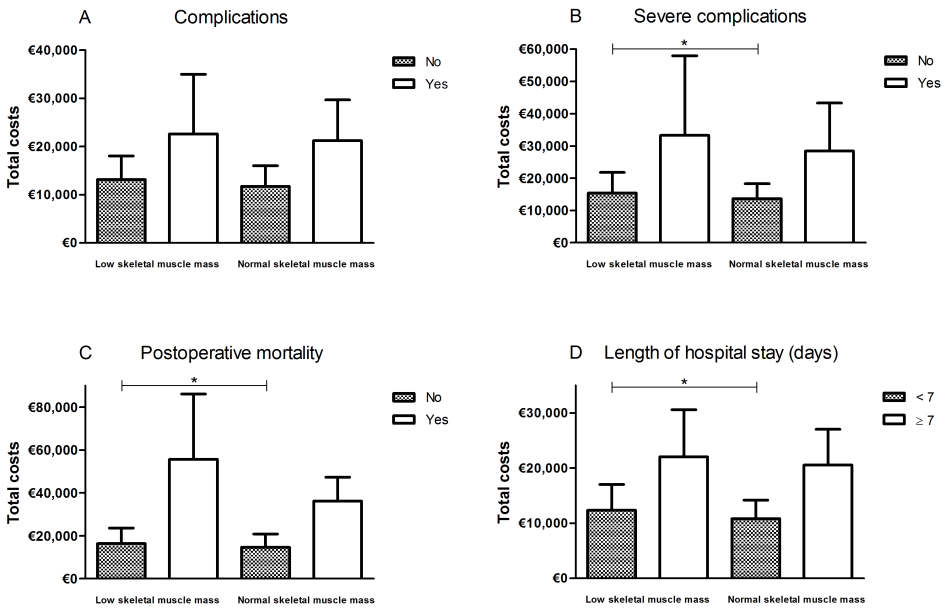

Figure 3. Total hospital costs stratified by the presence of low skeletal muscle mass per treatment outcome. * p<0.05, ** p<0.001

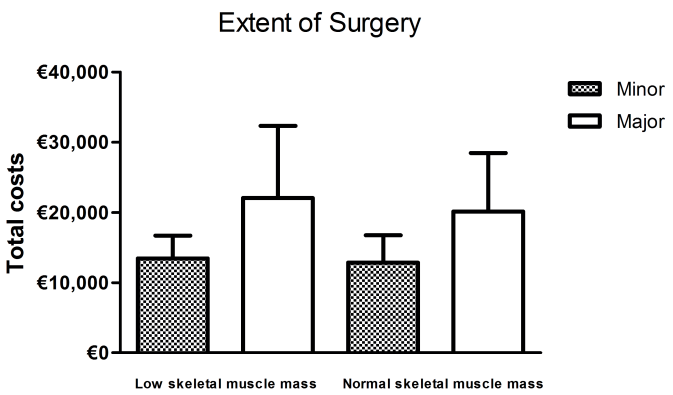

Figure 4. Total hospital costs stratified by the presence of low skeletal muscle mass per surgical treatment group. Major surgery included hepatic resections of at least two segments and a wedge resection or the resection of at least three hepatic segments, pancreatic surgery, and pelvic exenteration for locally advanced rectal cancer. Minor surgery included less than two hepatic segment resections and colorectal resections. * p<0.05

Multivariable linear regression analysis

In linear regression analysis, the presence of low skeletal muscle mass was independently associated with higher total hospital costs, after adjusting for the extent of surgery, sex, age, overweight, and ASA classification (table 4). Presence of low skeletal muscle mass resulted in a cost increase of €4,061 (95% confidence interval [CI] 809-7,312; p=0.015). When skeletal muscle index was used as a continuous parameter, an incremental increase in skeletal muscle index (cm^2/m^2) was associated with €278 (95% CI 32-524, p=0.027).

Table 4. Multivariable linear regression analysis for the total costs per patient.

	B, Euros	**Standard Error, Euros**	***p*-value**
Low skeletal muscle mass	4,061	1,654	0.015
Age (≥65 years)	484	1,648	0.769
Sex (male)	2,126	1,693	0.210
Overweight (BMI ≥25 kg/m^2)	-587	1,650	0.722
ASA classification (3 or 4)	7,205	2,044	<0.001
Extent of surgery (major surgery)	11,127	1,650	<0.001

Abbreviations: BMI, Body Mass Index; ASA, American Society of Anesthesiologists

DISCUSSION

In this study in a Western-European healthcare system among patients undergoing gastro-intestinal cancer surgery we found that the costs were €2,183 higher in patients with low skeletal muscle mass. Furthermore, total costs increased gradually across four levels of the skeletal muscle index for both men and women, which is in line with a previous study [7]. Finally, significant differences in costs in patients with low skeletal muscle mass compared with patients without low skeletal muscle mass were observed in patients undergoing major surgery, while this difference was not significant in patients undergoing minor surgery.

Advanced age is associated with both an increased risk of cancer and an increased risk of postoperative complications [25]. With increasing longevity and an increasingly aging population, the number of older cancer patients is steadily growing. About 58% of all cancers and 69% of cancer deaths occur in patients aged 65 years and older [26]. The number of elderly patients undergoing cancer surgery is also rapidly growing. For example, 50% of patients with colorectal cancer are 70 years or older [27]. Therefore, low skeletal muscle mass, resulting from both age-related sarcopenia and disease-related cachexia, may be a highly interesting parameter in an era of an increasingly aging population.

Besides increased costs, we found significantly more complications and a significantly longer hospital stay in patients with low skeletal muscle mass, which is in line with current literature [1]. Significant differences in hospital costs between patients with and without low skeletal muscle mass were particularly observed in patients who did not experience any (severe) postoperative complications, did not die during admission or within 30 days postoperatively, or did not have a prolonged hospital stay. This finding suggests that increased costs may be directly related to the occurrence of (severe) complications requiring an increased use of resources (e.g., prolonged (ICU) stay, laboratory tests, radiological examinations and radiological or surgical re-interventions), as previously described [28, 29]. This is underlined by the fact that we found a significant difference in patients undergoing major surgery, known for more complex care and relatively high postoperative complication rates, but not in patients undergoing minor surgery. However, based on our results these costs may preoperatively be predicted by CT-based skeletal muscle mass measurements. Skeletal muscle mass measurements are known for their great inter- and intra-observer agreement [30], while ASA classification, for example, is often considered a parameter that is not eligible for cancer patients

and subjective with great inter-rater inconsistency [31, 32]. Low skeletal muscle mass may thus be used as a parameter for case-mix corrections to compare treatment outcomes between centers.

To date, all studies performed to assess the association between low skeletal muscle mass and healthcare costs showed a positive association, favoring patients with normal skeletal muscle mass [7-9, 33, 34]. To our knowledge, this study is the first to show that low skeletal muscle mass may be a highly reliable measure to predict impaired outcome and increased hospital costs in surgical cancer patients in Western-European healthcare systems. Since European healthcare costs are increasingly rising and low skeletal muscle mass suggested to be a remediable condition, antagonizing skeletal muscle mass loss in cancer patients may be a powerful target to reduce hospital resource use and costs. Besides prehabilitation programs (i.e. physical exercise therapy combined with nutritional supplementation), promising results have been shown in animal studies [35]. Multiple phase II trials are currently being performed [36]. On the other hand, low skeletal muscle mass may be a measure of advanced disease, or even a final common pathway from cancer to death. Low skeletal muscle mass, as a measure of frailty and physical status of patients, may therefore also be added to case-mix correction models.

Some limitations of this study should be acknowledged. A number of costs during hospital admission (i.e. medication and feeding) may not have been recorded and consequently missed in the analyses. In addition, no data was available after discharge from our hospital. It has previously been shown that patients with low skeletal muscle mass experience a delayed recovery and are more likely not to rehabilitate at home [8, 37]. This may have led to an underestimation of the true total costs in patients with low skeletal muscle mass and thus an underestimation of the difference in costs between patients with and without low skeletal muscle mass. However, the total hospital costs are in line with a previous Dutch study investigating costs after major abdominal surgery in an academic center [28] and the study of Kirk *et al.* which showed that the greatest difference in costs occurs in the first 30 days after surgery [8]. Due to the retrospective design of this study, CT examinations have not been performed in identical scanners. A previous study, however, showed that fat area measurements (based on the same principle) on CT examinations performed with various scanners in individuals had excellent agreements, independent of the scanner used [38]. Some of the non-significant differences found may be explained by a limited sample size. Finally, we only investigated the association between skeletal muscle mass and healthcare costs, rather than sarcopenia, characterized by both skeletal muscle mass and function loss

[39], or cachexia, characterized by both skeletal muscle mass and body weight loss [40]. Although low skeletal muscle mass a hallmark parameter of both syndromes, future studies should confirm our results in patients with sarcopenia or cachexia.

In conclusion, total hospital costs are €4,061 higher in patients with low skeletal muscle mass compared with patients with normal skeletal muscle mass after correction for extent of surgery and ASA classification. The search for a treatment of low skeletal muscle mass might therefore lead to a reduction of complications and hospital costs in an era of incremental healthcare costs and an increasingly aging population. Future prospective studies regarding skeletal muscle mass should consider including total costs as outcome measure.

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REFERENCES

1. Levolger S, Van Vledder MG, Muslem R, Koek M, Niessen WJ, De Man RA, et al. Sarcopenia impairs survival in patients with potentially curable hepatocellular carcinoma. J Surg Oncol 2015; 112: 208-13.
2. van Vugt JL, Levolger S, de Bruin RW, van Rosmalen J, Metselaar HJ, IJzermans JN. Systematic Review and Meta-Analysis of the Impact of Computed Tomography-Assessed Skeletal Muscle Mass on Outcome in Patients Awaiting or Undergoing Liver Transplantation. Am J Transplant 2016; 16: 2277-92.
3. Shachar SS, Williams GR, Muss HB, Nishijima TF. Prognostic value of sarcopenia in adults with solid tumours: A meta-analysis and systematic review. Eur J Cancer 2016; 57: 58-67.
4. Huillard O, Mir O, Peyromaure M, Tlemsani C, Giroux J, Boudou-Rouquette P, et al. Sarcopenia and body mass index predict sunitinib-induced early dose-limiting toxicities in renal cancer patients. Br J Cancer 2013; 108: 1034-41.
5. Tan BH, Brammer K, Randhawa N, Welch NT, Parsons SL, James EJ, et al. Sarcopenia is associated with toxicity in patients undergoing neo-adjuvant chemotherapy for oesophago-gastric cancer. Eur J Surg Oncol 2015; 41: 333-8.
6. Antoun S, Baracos VE, Birdsell L, Escudier B, Sawyer MB. Low body mass index and sarcopenia associated with dose-limiting toxicity of sorafenib in patients with renal cell carcinoma. Annals of oncology : official journal of the European Society for Medical Oncology / ESMO 2010; 21: 1594-8.
7. Gani F, Buettner S, Margonis GA, Sasaki K, Wagner D, Kim Y, et al. Sarcopenia predicts costs among patients undergoing major abdominal operations. Surgery 2016.
8. Kirk PS, Friedman JF, Cron DC, Terjimanian MN, Wang SC, Campbell DA, et al. One-year postoperative resource utilization in sarcopenic patients. The Journal of surgical research 2015; 199: 51-5.
9. Sheetz KH, Waits SA, Terjimanian MN, Sullivan J, Campbell DA, Wang SC, et al. Cost of major surgery in the sarcopenic patient. Journal of the American College of Surgeons 2013; 217: 813-8.
10. Gornick ME, Eggers PW, Reilly TW, Mentnech RM, Fitterman LK, Kucken LE, et al. Effects of race and income on mortality and use of services among Medicare beneficiaries. The New England journal of medicine 1996; 335: 791-9.
11. Obama B. United States Health Care Reform: Progress to Date and Next Steps. JAMA 2016; 316: 525-32.
12. Kroneman M, Boerma W, van den Berg M, Groenewegen P, de Jong J, van Ginneken E. Netherlands: Health System Review. Health systems in transition 2016; 18: 1-240.

13. Healthcare in the Netherlands. Ministry of Public Health, Welfare and Sport; 2016; Available from: https://www.government.nl/documents/leaflets/2016/02/09/healthcare-in-the-netherlands.
14. Lalmahomed ZS, Coebergh van den Braak RR, Oomen MH, Arshad SP, Riegman PH, JN IJ, et al. Multicenter fresh frozen tissue sampling in colorectal cancer: does the quality meet the standards for state of the art biomarker research? Cell Tissue Bank 2017.
15. Buttner S, Lalmahomed ZS, Coebergh van den Braak RR, Hansen BE, Coene PP, Dekker JW, et al. Completeness of pathology reports in stage II colorectal cancer. Acta Chir Belg 2017; 117: 181-7.
16. Levolger S, van Vledder MG, Alberda WJ, Verhoef C, de Bruin RWF, JNM IJ, et al. Muscle wasting and survival following pre-operative chemoradiotherapy for locally advanced rectal carcinoma. Clin Nutr 2017.
17. Levolger S, van Vledder MG, Muslem R, Koek M, Niessen WJ, de Man RA, et al. Sarcopenia impairs survival in patients with potentially curable hepatocellular carcinoma. J Surg Oncol 2015; 112: 208-13.
18. van Vledder MG, Levolger S, Ayez N, Verhoef C, Tran TC, IJzermans JN. Body composition and outcome in patients undergoing resection of colorectal liver metastases. Br J Surg 2012; 99: 550-7.
19. Buettner S, van Vugt JL, Gaspersz MP, Coelen RJ, Roos E, Labeur T, et al. Survival after Resection of Perihilar Cholangiocarcinoma in Patients with Lymph Node Metastases. HPB (Oxford) 2017.
20. Morak MJ, van der Gaast A, Incrocci L, van Dekken H, Hermans JJ, Jeekel J, et al. Adjuvant intra-arterial chemotherapy and radiotherapy versus surgery alone in resectable pancreatic and periampullary cancer: a prospective randomized controlled trial. Annals of surgery 2008; 248: 1031-41.
21. Dripps RD, Lamont A, Eckenhoff JE. The role of anesthesia in surgical mortality. JAMA 1961; 178: 261-6.
22. Ogden CL, Carroll MD, Kit BK, Flegal KM. Prevalence of obesity and trends in body mass index among US children and adolescents, 1999-2010. Jama 2012; 307: 483-90.
23. Dindo D, Demartines N, Clavien PA. Classification of surgical complications: a new proposal with evaluation in a cohort of 6336 patients and results of a survey. Annals of surgery 2004; 240: 205-13.
24. Martin L, Birdsell L, Macdonald N, Reiman T, Clandinin MT, McCargar LJ, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol 2013; 31: 1539-47.
25. DePinho RA. The age of cancer. Nature 2000; 408: 248-54.
26. Coebergh JW. Epidemiology in Europe. Eur J Cancer 2001; 37 Suppl 7: S217-27.

27. Snijders HS, Wouters MW, van Leersum NJ, Kolfschoten NE, Henneman D, de Vries AC, et al. Meta-analysis of the risk for anastomotic leakage, the postoperative mortality caused by leakage in relation to the overall postoperative mortality. Eur J Surg Oncol 2012; 38: 1013-9.
28. Straatman J, Cuesta MA, de Lange-de Klerk ES, van der Peet DL. Hospital cost-analysis of complications after major abdominal surgery. Dig Surg 2015; 32: 150-6.
29. Vonlanthen R, Slankamenac K, Breitenstein S, Puhan MA, Muller MK, Hahnloser D, et al. The impact of complications on costs of major surgical procedures: a cost analysis of 1200 patients. Annals of surgery 2011; 254: 907-13.
30. van Vugt JL, Levolger S, Gharbharan A, Koek M, Niessen WJ, Burger JW, et al. A comparative study of software programmes for cross-sectional skeletal muscle and adipose tissue measurements on abdominal computed tomography scans of rectal cancer patients. J Cachexia Sarcopenia Muscle 2017; 8: 285-97.
31. Mak PH, Campbell RC, Irwin MG, American Society of A. The ASA Physical Status Classification: inter-observer consistency. American Society of Anesthesiologists. Anaesth Intensive Care 2002; 30: 633-40.
32. Makary MA, Segev DL, Pronovost PJ, Syin D, Bandeen-Roche K, Patel P, et al. Frailty as a predictor of surgical outcomes in older patients. Journal of the American College of Surgeons 2010; 210: 901-8.
33. Wang SL, Zhuang CL, Huang DD, Pang WY, Lou N, Chen FF, et al. Sarcopenia Adversely Impacts Postoperative Clinical Outcomes Following Gastrectomy in Patients with Gastric Cancer: A Prospective Study. Ann Surg Oncol 2016; 23: 556-64.
34. Dunn MA, Josbeno DA, Tevar AD, Rachakonda V, Ganesh SR, Schmotzer AR, et al. Frailty as Tested by Gait Speed is an Independent Risk Factor for Cirrhosis Complications that Require Hospitalization. Am J Gastroenterol 2016.
35. Zhou X, Wang JL, Lu J, Song Y, Kwak KS, Jiao Q, et al. Reversal of cancer cachexia and muscle wasting by ActRIIB antagonism leads to prolonged survival. Cell 2010; 142: 531-43.
36. Dingemans AM, de Vos-Geelen J, Langen R, Schols AM. Phase II drugs that are currently in development for the treatment of cachexia. Expert Opin Investig Drugs 2014; 23: 1655-69.
37. Lieffers JR, Bathe OF, Fassbender K, Winget M, Baracos VE. Sarcopenia is associated with postoperative infection and delayed recovery from colorectal cancer resection surgery. Br J Cancer 2012; 107: 931-6.
38. Yoshizumi T, Nakamura T, Yamane M, Islam AH, Menju M, Yamasaki K, et al. Abdominal fat: standardized technique for measurement at CT. Radiology 1999; 211: 283-6.
39. Rolland Y, Abellan van Kan G, Gillette-Guyonnet S, Vellas B. Cachexia versus sarcopenia. Current opinion in clinical nutrition and metabolic care 2011; 14: 15-21.
40. Fearon K, Strasser F, Anker SD, Bosaeus I, Bruera E, Fainsinger RL, et al. Definition and classification of cancer cachexia: an international consensus. Lancet Oncol 2011; 12: 489-95.

CHAPTER 13

Low Skeletal Muscle Mass is Associated with Increased Hospital Costs in Patients with Cirrhosis Listed for Liver Transplantation – A Retrospective Study

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ABSTRACT

Low skeletal muscle mass (sarcopenia) is associated with increased morbidity and mortality in liver transplant candidates. We investigated the association between sarcopenia and hospital costs in patients listed for liver transplantation. Consecutive patients with cirrhosis listed for liver transplantation between 2007-2014 in a Eurotransplant center were identified. The skeletal muscle index ([SMI], cm^2/m^2) was measured on CT performed within 90 days from waiting list placement. The lowest sex-specific quartile represented patients with sarcopenia. In total, 224 patients were included. Median time on the waiting list was 170 (IQR 47-306) days and median MELD-score was 16 (IQR 11-20). The median total hospital costs in patients with sarcopenia were €11,294 (IQR 3,570-46,469) compared with €6,878 (IQR 1,305-20,683) in patients without sarcopenia (p=0.008). In multivariable regression analysis, an incremental increase in SMI was significantly associated with a decrease in total costs (€455 per incremental SMI, 95%CI 11-900, p=0.045), independent of the total time on the waiting list. In conclusion, sarcopenia is independently associated with increased health-related costs for patients on the waiting list for liver transplantation. Optimizing skeletal muscle mass may therefore lead to a decrease in hospital expenditure, in addition to greater health benefit for the transplant candidate.

INTRODUCTION

Liver transplantation is the only curative treatment for patients with end-stage liver disease [1]. The 1-year and 3-year survival rates of patients who undergo orthotopic liver transplantation in Europe and the United States are around 85% and 80%, respectively [1]. While allocation of donor organs is based on the Model for End-stage Liver Disease (MELD) score [2, 3], which measures liver function, patients on the waiting list are at increased risk for major morbidity and mortality, particularly due to infections [1, 4-6]. Indeed, hospital admissions in patients with end-stage liver disease occur frequently and are costly [7-9].

One of the factors related to hospital admissions is frailty, which is defined as the increased vulnerability to stressors due to reduced physiological reserves. Frailty is a known risk factor for adverse outcome in cirrhosis and liver transplant patients [10-13]. Sarcopenia, defined as the involuntary loss of skeletal muscle mass and function, is part of the frailty syndrome and highly prevalent among patients with end-stage liver disease [14]. In patients with cirrhosis, low skeletal muscle mass is associated with increased mortality on the liver transplantation waiting list and post-transplant morbidity and mortality, independently of well-established predictors such as the MELD score [14, 15]. Sarcopenia has also been associated with higher healthcare costs in abdominal cancer patients undergoing surgery [16-18]. To date, only one study from the United States described the association between gait speed, as a measure of frailty, and increased hospital costs in patients with cirrhosis [11]. However, generalizability of these data is limited because, as a consequence of income inequality, great differences exist between the United States and Western Europe regarding healthcare accessibility [19-21].

The primary objective of this study, therefore, was to investigate the association between skeletal muscle mass and hospital costs in patients with cirrhosis listed for liver transplantation in a European transplant center. A secondary objective was to assess the association between skeletal muscle mass and total hospital costs during admission for liver transplantation in the subgroup of patients who eventually underwent liver transplantation.

METHODS

Patients and data acquisition

All consecutive patients who were listed for liver transplantation from January 2007 to December 2014 at Erasmus MC University Medical Center were identified using the Eurotransplant registry [22]. Patients listed for reasons other than cirrhosis (n=30), patients with acute liver failure/listed with high urgency (n=58), patients undergoing re-transplantation (n=58), and those removed because of clinical improvement (n=9) or other reasons such as patient preferences or substance abuse (n=5) were excluded. The following parameters were collected at the moment of liver transplantation screening: sex, age, body height and weight, etiology of liver disease, blood group, MELD score, and the occurrence of complications (i.e. ascites, spontaneous bacterial peritonitis, hepatic encephalopathy, or variceal bleeding) before listing. All hospital admissions (including one-day admissions) with corresponding indication were recorded and the cumulative days of hospital stay were calculated. The indication for hospital admission was scored as follows: decompensated cirrhosis, infection, scheduled intervention (e.g., transarterial chemoembolization (TACE), radiofrequency ablation (RFA), endoscopic retrograde cholangio-pancreatography (ERCP), coloscopy, biopsy), other, or unknown.

The endpoint of the study was reached when patients underwent liver transplantation, were removed from the waiting list (due to clinical deterioration), or died on the waiting list. Patients who were removed from the waiting list because of clinical improvement or who were still on the waiting list at December 31, 2016, were excluded. All patients with hepatocellular carcinoma (HCC) were transplanted within the Milan criteria [23]. Patients with HCC with disease progression beyond the Milan criteria were removed from the waiting list and considered as clinically deteriorated. In the study period, no prehabilitation program was conducted.

In patients who underwent liver transplantation, the cumulative length of hospital stay (LOS) was calculated as the sum of the index admission and all readmissions within 30 days of discharge. The Institutional Review Board approved the study and a waiver for informed consent was granted.

Skeletal muscle mass measurements

The cross-sectional skeletal muscle area (cm^2) was measured on contrast-enhanced (portal-venous phase) abdominal computed tomography (CT) at the level of the third lumbar vertebra (L3) and adjusted for patients' height squared, as previously described (figure 1) [24]. This resulted in the skeletal muscle index (cm^2/m^2), a measure strongly

correlated with total body skeletal muscle mass [25]. Established cut-off values take body mass index (BMI) into account [26], which is known to be inaccurate in patients with liver failure due to ascites and peripheral edema. Consequently, sex-specific skeletal muscle mass quartiles were created. Patients in the lowest sex-specific quartile were considered to have sarcopenia. CT scans closest to the date of listing, but within 90 days from the listing date, were used for analyses.

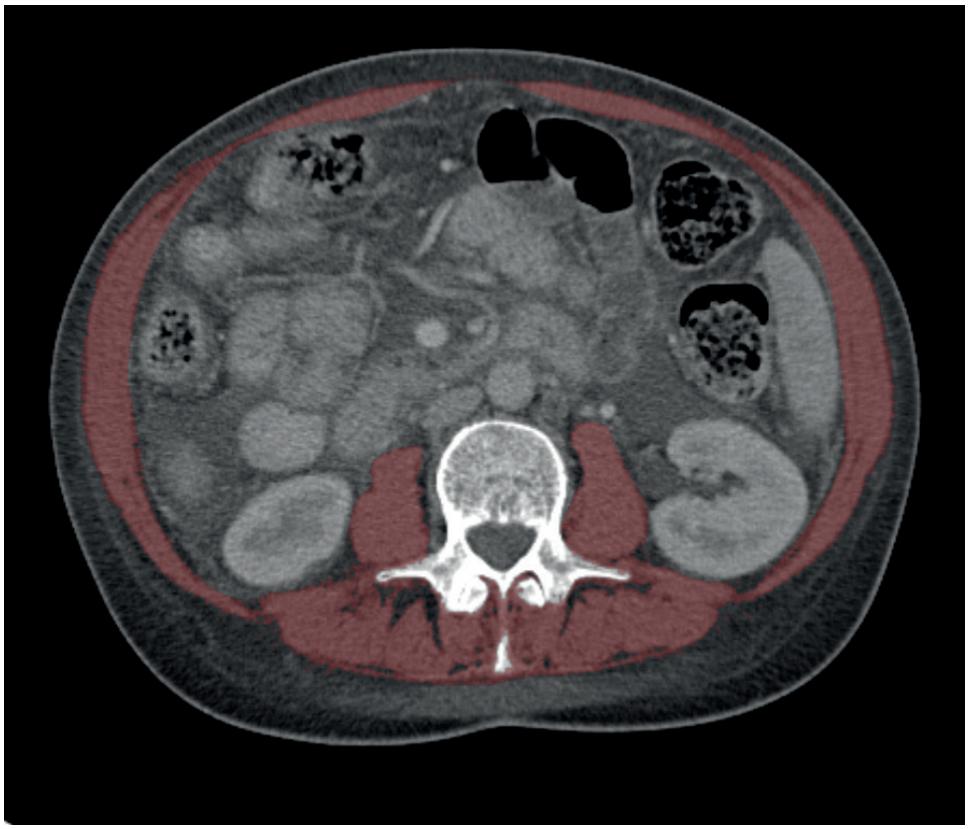

Figure 1. Cross-sectional skeletal muscle mass measurement.Example of a measurement of skeletal muscle mass on CT. The cross-sectional skeletal muscle area (129.74 cm^2) is depicted of a 60-year-old female with a body mass index of 22.1 kg/m^2. With a body height of 1.74 meters this resulted in a skeletal muscle index of 42.9 cm^2/m^2. Consequently, this patient was considered not to have sarcopenia.

Cost analyses

All hospital costs (i.e. both clinical and outpatient department costs) that were made during the period that patients were listed for liver transplantation (i.e. from the date of listing to the endpoint, excluding hospitalization for liver transplantation) were included, as previously described. Costs for medication were not included. In patients who underwent liver transplantation, total hospital costs during index admission (including the day of liver transplantation) for the liver transplantation and during readmission(s) within 30 days after discharge from the index admission were also collected. In these transplanted patients, the grand total was calculated by adding the total hospital costs during waiting list placement and total hospital costs during hospital admission for liver transplantation.

Costs were extracted from the hospital's electronic accounting system. Total costs were calculated by the sum of all unit cost prices. Financial data was limited to hospital expenditure and did not include costs made outside our center. Adjustment for inflation was performed by indexing all cost prices to the year 2015 according to data of the Dutch Healthcare Authority. All financial data are reported in Euros (€).

Statistical analyses

Categorical data are reported as counts with percentages. Continuous data are reported as median with interquartile range (IQR) or mean with standard deviation (SD), depending on their distribution. The Chi-square test was used to compare categorical data, whereas the Mann-Whitney-U test was used to compare hospital costs between patients with and without sarcopenia. A multivariable linear regression analysis was performed to investigate the association of an incremental skeletal muscle index with total hospital costs after correction for possible confounding and clinically relevant factors. Sex was added to the model to adjust for differences in skeletal muscle mass per gender. Sex-specific skeletal muscle mass quartiles were compared using the Kruskal-Wallis test. Subgroup analyses were performed for the presence of HCC. Two-sided p-values <0.05 were considered statistically significant. All analyses were performed using SPSS for Windows (IBM Corp., Armonk, NY, USA), version 22.

RESULTS

Patients

In total, 362 patients with cirrhosis were listed for liver transplantation, of whom 224 (61.9%) patients were eligible for the study (figure 2). Baseline characteristics are shown in table 1. Of these patients, 149 (66.5%) were male and 75 (33.5%) had concomitant HCC. Baseline characteristics and total hospital costs did not significantly differ between the included and excluded patients (data not shown). Baseline characteristics and outcome (i.e. total costs) did not significantly differ between in- and excluded patients.

In total, 165 (73.7%) patients eventually underwent liver transplantation. The remaining patients were removed from the waiting list due to infections (12.9%), rapid clinical deterioration with decompensated cirrhosis (2.7%), progression of HCC beyond the Milan criteria (8.5%), diagnosis of other malignancies (1.3%) or cardiopulmonary decompensation (0.9%).

Hospital costs

The median total hospital costs across the entire study cohort were €7,761 (IQR 1,630-23,954), corresponding to €44 (IQR 12-164) per day on the waiting list. The median total hospital costs were significantly lower in patients who eventually underwent liver transplantation compared with patients who were removed from the waiting list (i.e. due to mortality, clinical deterioration, progression of HCC beyond the Milan criteria, or other malignancies).Furthermore, costs were significantly higher in patients without HCC compared with patients with HCC (table 2).

Skeletal muscle mass and total hospital costs during the waiting list period

The median time between CT and waiting list placement was 30 (IQR 17-51) days. The median skeletal muscle index was 50.4 cm^2/m^2 (IQR 44.1-55.0) for males and 41.8 cm^2/m^2 (IQR 37.9-46.5) for females ($p<0.001$).

Total hospital costs decreased per incremental increase in SMI sex-specific quartile (Figures 3a and 3b). The median total hospital costs in patients with sarcopenia were €11,294 (IQR 3,570-46,469) compared with €6,878 (IQR 1,305-20,683) in patients without sarcopenia (p=0.008, table 2). This corresponds to €68 (IQR 16-503) per day on the waiting list in patients with sarcopenia compared with €40 (IQR 10-108) in patients without sarcopenia (p=0.013).

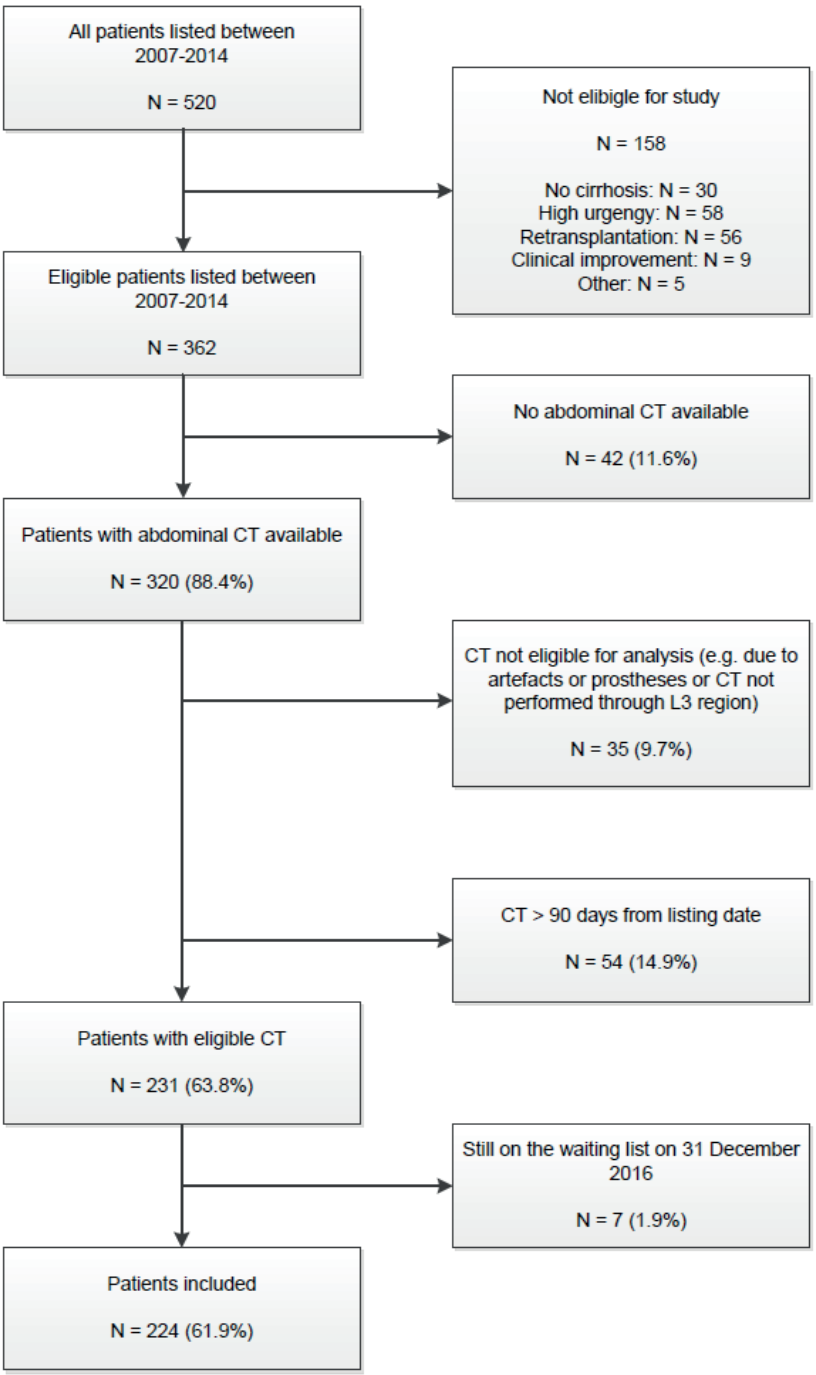

Figure 2. Inclusion flowchart.

Multivariable linear regression analysis on costs during the waiting list period

Adjusted for age at the moment of listing, sex, MELD score at the moment of listing, complications before listing, presence of malignancy (i.e. HCC or cholangiocarcinoma), and total time on the waiting list, an incremental increase in SMI was significantly associated with a decrease in total hospital costs (€455 per incremental increase in SMI, 95% CI 11-900, p=0.045), independent of the total time on the waiting list (table 3).

Subgroup analyses in patients with and without HCC

Because patients without HCC had significantly higher total hospital costs compared with patients with HCC and a significantly higher number of HCC was observed in patients without sarcopenia compared with patients with sarcopenia (39.6% versus 14.5%, p<0.001), subgroup analyses in patients with and without HCC were performed. Significantly more males then females presented with HCC (77.6% versus 22.4%, p=0.012). The median MELD score was significantly lower in patients with HCC compared with patients without HCC (10 [IQR 8-12] versus 19 [IQR 16-22], p<0.001).

Patients without HCC and sarcopenia had significantly higher total costs compared with patients without HCC without sarcopenia (€19,586 [IQR 3,573-52,406] versus €7,644 [IQR 1,462-28,074], p=0.023), whereas no difference was found between HCC patients with and without sarcopenia (€4,610 [IQR 1,792-10,243] versus €5,001 [IQR 1,112-12,209], p=0.933). In a multivariable linear regression model in patients without HCC, an incremental increase in SMI was associated with decreased total hospital costs (€692 per incremental increase in SMI, 95% CI 77-1,306, p=0.028), independently of total time on the waiting list (€29 per day, 95% CI 12-46, p=0.001) (Supplementary table 1).

Skeletal muscle mass and total hospital costs in transplanted patients

The median time on the waiting list for the 165 patients (73.7%) who eventually underwent liver transplantation was 176 (IQR 51-306) days. The median time between CT and liver transplantation was 213 (IQR 90-343) days. Waiting time did not significantly differ between patients with and without sarcopenia (168 [IQR 39-301] versus 205 [IQR 52-311] days, p=0.597), although patients with sarcopenia had a significantly higher MELD score on the moment of waiting list placement (18 [IQR 13-21] versus 15 [IQR 10-19], p=0.044), and fewer patients had HCC (18.2% versus 39.7%, p=0.010) compared with patients without sarcopenia. The cumulative post-transplant LOS did not significantly differ between patients with and without sarcopenia (26 (IQR 15-36] versus 20 [IQR 15-32] days, p=0.124).

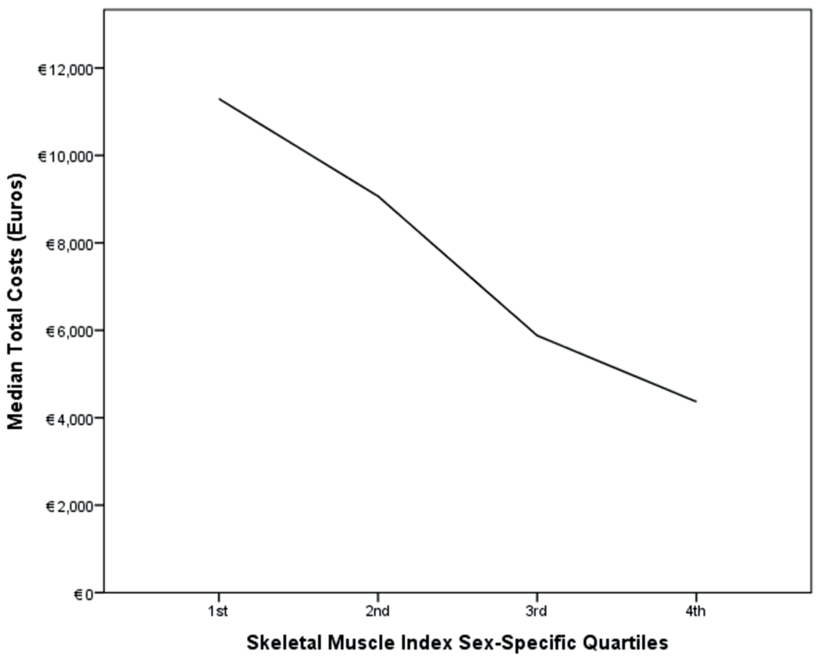

Figure 3a. Total hospital costs by skeletal muscle mass in sex-specific quartiles. The total hospital costs significantly decreased per sex-specific skeletal muscle mass quartile from a median of €11,294 (IQR 3,570-46,469) in the first quartile, €9,066 (IQR 1,515-26,648) in the second quartile, €5,781 (IQR 910-19,928) in the third quartile, to €4,366 (IQR 1,550-17,490) in the fourth quartile (p=0.026).

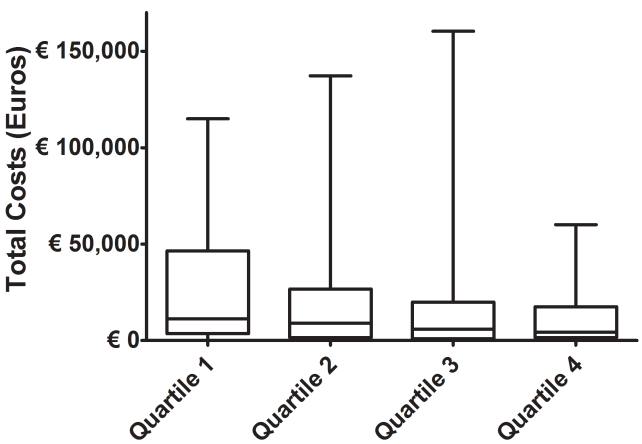

Figure 3b. The total hospital costs presented in a box-plot.l

Table 1. Baseline characteristics.

	Sarcopenia (n=55)	No sarcopenia (n=169)	p-value
Sex (male)	37 (67.3)	112 (66.3)	0.891
Age (years)	56 (48-62)	56 (49-61)	0.954
BMI (kg/m^2)	23.1 (21.6-25.1)	26.3 (23.6-29.4)	<0.001
Primary etiology of cirrhosis			
Alcoholic	9 (16.4)	19 (11.2)	0.012
Hepatitis B virus	3 (5.5)	4 (2.4)	
Hepatitis C virus	3 (5.5)	13 (7.7)	
PSC/PBC	21 (38.2)	44 (26.0)	
HCC	8 (14.5)	67 (39.6)	
Cholangiocarcinoma	0 (0.0)	1 (0.6)	
NASH	5 (9.1)	2 (1.2)	
Cryptogenic	2 (3.6)	7 (4.1)	
Auto-immune hepatitis	1 (1.8)	4 (2.4)	
Other	3 (5.5)	8 (4.7)	
Blood type			
O	29 (52.7)	67 (39.6)	0.264
A	19 (34.5)	67 (39.6)	
B	6 (10.9)	24 (14.2)	
AB	1 (1.8)	11 (6.5)	
MELD score	18 (15-21)	15 (11-20)	0.012
Complications before waiting list placement			
Any	45 (81.8)	112 (66.3)	0.029
Ascites	43 (78.2)	99 (58.6)	0.009
Spontaneous bacterial peritonitis	12 (21.8)	24 (14.2)	0.182
Hepatic encephalopathy	20 (36.4)	41 (24.3)	0.080
Esophageal variceal bleeding	16 (29.1)	42 (24.9)	0.533
Median days on the waiting list	165 (32-374)	170 (51-304)	0.755

Abbreviations: BMI, Body Mass Index; PSC, Primary Sclerosing Cholangitis; PBC, Primary Biliary Cirrhosis; HCC, Hepatocellular Carcinoma; NASH, Non-alcoholic Steatohepatitis; MELD, Model for End-stage Liver Disease.

Table 2. Indications for hospitalization during the waiting list period (n=194).

	n	%
Decompensation of cirrhosis		
Clinical deterioration of liver function	3	1.5
Ascites	26	13.4
Hepatic encephalopathy (+ infection)	10 (5)	5.2 (2.6)
Infection	31	16.0
SBP (with hepatic encephalopathy)	5 (1)	2.6 (0.5)
Scheduled intervention	47	24.2
Other	62	32.0
Unknown	4	2.1

Abbreviations: SBP, Spontaneous Bacterial Peritonitis

The median total hospital costs during the admission for liver transplantation of the entire cohort were €77,074 (IQR 54,410-98,505). These did not significantly differ between patients with and without sarcopenia (€81,569 [IQR 59,233-108,190] versus €74,612 [IQR 52,899-93,632], p=0.202). The grand total was €86,412 (IQR 62,478-113,791). This was significantly higher in patients with sarcopenia (€98,703 [IQR 75,909-121,071]) compared with patients without sarcopenia (€81,173 [IQR 58,961-110,258]), p=0.030.

Hospitalization and hospital costs

In total, 52 (23.2%) patients were admitted to the hospital during the waiting list period minimal once, accounting for 194 hospital admissions. In these patients, the median number of hospitalizations was 2 (IQR 1-3) with a median stay of 6 (IQR 2-14) days. The most frequent indication for hospitalization was a scheduled intervention (n=47, 24.2%), followed by infection (n=42, 21.6%) (table 4). Twenty-six (50.0%) patients were hospitalized because of decompensated disease at least once, with a median of 2 (IQR 1-3) hospital admissions for decompensated disease. Patients who were admitted during the waiting list period had significantly higher hospital costs (€16,799 [IQR 9,046-29,064] versus €4,396 [IQR 1,049-20,043], p<0.001). Although a precise estimation could not be made, this would mean that an average day in the hospital costs a total of €1,034. Number of admissions (p=0.640) and cumulative length of hospitalization (p=0.609), however, did not differ between patients with and without sarcopenia. A not statistically significant higher proportion of patients with sarcopenia was admitted because of liver decompensation (n=9, 64.3%) compared with patients without sarcopenia (n=17, 44.7%), p=0.211. These patients also showed a higher number of hospital admissions for decompensated disease (3 [IQR 0-2] versus 1 [IQR 0-4], p=0.150).

In patients without sarcopenia, those who were admitted had significantly higher hospital costs compared with those not admitted (€17,529 [IQR 10,088-27,989] versus €3,503 [IQR 979-16,267], p<0.001). However, in patients with sarcopenia, those who were admitted had comparable hospital costs compared with those not admitted (€16,087 [IQR 7,878-53,319] versus €10,746 [IQR 2,227-45,602], p=0.324).

Table 3. Multivariable linear regression analysis for the total hospital costs per patient during waiting list placement (n=224).

	Euros per unit	Standard Error, Euros	p-value
Skeletal muscle index (per cm^2/m^2)	-455	226	0.045
Sex (female versus male)	-3,146	3,890	0.420
Age at moment of listing (per year)	18	163	0.911
MELD score at moment of listing (per point)	507	380	0.184
Complications before listing (yes versus no)	2,134	4,036	0.598
Malignancy (HCC/Cholangiocarcinoma versus other indication)	-4,305	4,741	0.365
Time on the waiting list (per day)	30	7	<0.001

Abbreviations: MELD, Model for End-stage Liver Disease; HCC, Hepatocellular Carcinoma

Table 4. Indications for hospitalization during the waiting list period (n=194).

	n	%
Decompensation of cirrhosis		
Clinical deterioration of liver function	3	1.5
Ascites	26	13.4
Hepatic encephalopathy (+ infection)	10 (5)	5.2 (2.6)
Infection	31	16.0
SBP (with hepatic encephalopathy)	5 (1)	2.6 (0.5)
Scheduled intervention	47	24.2
Other	62	32.0
Unknown	4	2.1

Abbreviations: SBP, Spontaneous Bacterial Peritonitis

DISCUSSION

To the best of our knowledge, this is the first study to describe that health care costs in patients with cirrhosis and sarcopenia listed for liver transplantation are higher, and in our case involve almost €4,500 more, than patients without sarcopenia. In patients who eventually underwent liver transplantation, the difference in total costs (i.e. the sum of the hospital costs during waiting list placement and during the admission for transplantation) was even higher, over €17,000.

Frailty has previously been investigated in cirrhosis patients by Dunn *et al.* and Sinclair *et al.*, and their conclusion is in line with our findings on sarcopenia. Frailty, a measure for contractile function and balance, was found to be an independent risk factor for cirrhosis complications needing hospitalization [11, 27] and increased hospital costs [11]. The waiting list period offers a window of opportunity to improve functional status and skeletal muscle mass. Suggested regimens in patients with cirrhosis may consist of the use of proteins with low ammoniagenic potential, leucine enriched amino acid supplementation, long-term ammonia lowering strategies and a combination of resistance and endurance exercise to increase muscle mass and function [28]. Reversing or halting skeletal muscle wasting may lead to decreased costs on the waiting list.

We found significantly lower hospital costs in patients with HCC compared with patients without HCC. The significantly lower MELD score in patients with HCC compared with patients without HCC may explain this difference. After all, the lower median MELD score indicates less severity of the liver disease in patients with HCC. In addition to cancer [29] and age [30], liver disease itself is an important cause of skeletal muscle depletion [28]. Not only alterations in food intake, hypermetabolism, amino acid profiles, endotoxemia, accelerated starvation and decreased mobility lead to liver disease induced skeletal muscle depletion, but recent findings also indicate hyper ammonia as a mediator in the liver-muscle axis [28].

Although the association between sarcopenia and hospital expenditure is strong, we do not believe this to be a causal relationship. Instead, we believe that cirrhotic frail patients or those with sarcopenia are at increased risk for morbidity and mortality due to clinical and subclinical sequelae [12] and have increased (re)admission rates [8, 9, 11] which eventually lead to increased hospital costs [11]. Although we did not find differences in hospital admissions in general between patients with and without sarcopenia, we found a not statistically significant difference in the proportion of hospital admissions due to liver decompensation in favor of patients without sarcopenia. The low number of patients may have led to a type II error.

The significantly lower prevalence of sarcopenia among patients with HCC seems to be in contrast with previous studies describing a high prevalence of sarcopenia among patients with HCC [29, 31]. However, our study cohort consisted of patients within the Milan criteria only [23] and consequently the tumor load was limited. Furthermore, this difference may also be explained by the use of different cut-off values instead of continuous SMI in those studies [14]. Many previous studies of liver transplant patients used cut-off values based on body mass index (BMI) or body surface area (BSA). In our opinion this is a suboptimal measurement, as both measures are calculated using body weight in patients with ascites [32-35]. In a large series of Japanese patients with HCC (n=1,257), using cut-off values to predict mortality using optimal stratification in their patient cohort, a prevalence of low skeletal muscle mass of only 11.1% was found [31].

A statistically non-significant difference of €6,957 in hospital costs during the admission for liver transplantation, and a statistically significant difference of €17,530 in the grand total, favoring patients without sarcopenia was found. Since this was not the primary objective of the study, skeletal muscle mass was not measured on the CT closest to transplantation. Consequently, the median time interval between CT and liver transplantation was 208 days and the subgroup was relatively small (n=166). As patients may lose significant amounts of skeletal muscle mass during the waiting list period [13], these results should be interpreted with caution and should be validated in a future study with a smaller interval between CT and transplantation. The waiting list period may be used to halt or reverse skeletal muscle wasting. Currently, promising results have been shown in animal studies and multiple human phase II trials are being performed [36, 37].

There are no widely accepted cut-off values to classify patients as having sarcopenia yet. The most commonly used cut-off values are those of Martin and colleagues, established in a cohort of cancer patients [26]. Recently, cut-off values for patients with end-stage liver disease have been proposed in a North-American population, which have not been validated yet [38]. Due to differences between the American en European population, we chose to use our own cut-off values to exemplify cost differences between patients with low and high skeletal muscle mass. However, the independent association between skeletal muscle mass and hospital expenditure was shown using the skeletal muscle index (cm^2/m^2) as a continuous measure.

Although sarcopenia is a subject of interest in patients with liver disease, we are the first to show the actual costs involved alongside this comorbidity. However, there are some limitations in this study that need to be addressed. Firstly, we were, not able to include healthcare costs made outside the hospital. However, the median hospital costs

during the waiting list period (€ 7,761) were comparable with a previous German study (€ 6,294) [39]. Furthermore, we may have missed some costs and these results should, therefore, be considered as estimates. Although the current results may consequently be an underestimation of the real costs, one may expect that sarcopenia is associated with increased resource utilization after hospital discharge. We therefore believe that the difference between patients with and without sarcopenia might be underestimated rather than overestimated. Secondly, selection bias may have occurred due to the retrospective design of the study. However, all consecutive patients listed for liver transplantation were identified. Although a substantial part of patients listed for liver transplantation was excluded, significant differences were not found between baseline characteristics and outcome. Consequently, selection bias seems highly unlikeable. Thirdly, we only measured skeletal muscle mass and did not assess muscle function. Lastly, we were not able to monitor skeletal muscle wasting over time because consecutive CT examinations were not routinely performed.

In conclusion, sarcopenia is independently associated with higher hospital costs during waiting list placement of liver transplant candidates, as well as with higher total hospital costs (i.e. during waiting list placement and the admission for transplantation) in patients undergoing liver transplantation. Optimizing patients' skeletal muscle mass may therefore lead to a decrease in hospital expenditure. The differences in costs justify the efforts and the use of resources to explore therapies and treatments to reduce or stop skeletal muscle wasting in patients with end-stage liver disease. Furthermore, it underlines that low skeletal muscle mass may be used as a parameter for case-mix comparisons and corrections.

REFERENCES

1. Cardenas A, Gines P. Management of patients with cirrhosis awaiting liver transplantation. Gut 2011; 60: 412-21.
2. Freeman RB, Jr., Wiesner RH, Harper A, McDiarmid SV, Lake J, Edwards E, et al. The new liver allocation system: moving toward evidence-based transplantation policy. Liver Transpl 2002; 8: 851-8.
3. Kamath PS, Kim WR, Advanced Liver Disease Study G. The model for end-stage liver disease (MELD). Hepatology 2007; 45: 797-805.
4. Jalan R, Fernandez J, Wiest R, Schnabl B, Moreau R, Angeli P, et al. Bacterial infections in cirrhosis: a position statement based on the EASL Special Conference 2013. J Hepatol 2014; 60: 1310-24.
5. Arvaniti V, D'Amico G, Fede G, Manousou P, Tsochatzis E, Pleguezuelo M, et al. Infections in patients with cirrhosis increase mortality four-fold and should be used in determining prognosis. Gastroenterology 2010; 139: 1246-56, 56 e1-5.
6. Alferink LJ, Oey RC, Hansen BE, Polak WG, van Buuren HR, de Man RA, et al. The impact of infections on delisting patients from the liver transplantation waiting list. Transpl Int 2017; 30: 807-16.
7. Volk ML, Tocco RS, Bazick J, Rakoski MO, Lok AS. Hospital readmissions among patients with decompensated cirrhosis. Am J Gastroenterol 2012; 107: 247-52.
8. Bajaj JS, Reddy KR, Tandon P, Wong F, Kamath PS, Garcia-Tsao G, et al. The 3-month readmission rate remains unacceptably high in a large North American cohort of patients with cirrhosis. Hepatology 2016; 64: 200-8.
9. Berman K, Tandra S, Forssell K, Vuppalanchi R, Burton JR, Jr., Nguyen J, et al. Incidence and predictors of 30-day readmission among patients hospitalized for advanced liver disease. Clin Gastroenterol Hepatol 2011; 9: 254-9.
10. Makary MA, Segev DL, Pronovost PJ, Syin D, Bandeen-Roche K, Patel P, et al. Frailty as a predictor of surgical outcomes in older patients. Journal of the American College of Surgeons 2010; 210: 901-8.
11. Dunn MA, Josbeno DA, Tevar AD, Rachakonda V, Ganesh SR, Schmotzer AR, et al. Frailty as Tested by Gait Speed is an Independent Risk Factor for Cirrhosis Complications that Require Hospitalization. Am J Gastroenterol 2016; 111: 1768-75.
12. Lai JC, Feng S, Terrault NA, Lizaola B, Hayssen H, Covinsky K. Frailty predicts waitlist mortality in liver transplant candidates. Am J Transplant 2014; 14: 1870-9.
13. Lai JC, Dodge JL, Sen S, Covinsky K, Feng S. Functional decline in patients with cirrhosis awaiting liver transplantation: Results from the functional assessment in liver transplantation (FrAILT) study. Hepatology 2016; 63: 574-80.

14. van Vugt JL, Levolger S, de Bruin RW, van Rosmalen J, Metselaar HJ, IJzermans JN. Systematic Review and Meta-Analysis of the Impact of Computed Tomography-Assessed Skeletal Muscle Mass on Outcome in Patients Awaiting or Undergoing Liver Transplantation. Am J Transplant 2016; 16: 2277-92.
15. Durand F, Buyse S, Francoz C, Laouenan C, Bruno O, Belghiti J, et al. Prognostic value of muscle atrophy in cirrhosis using psoas muscle thickness on computed tomography. J Hepatol 2014; 60: 1151-7.
16. Gani F, Buettner S, Margonis GA, Sasaki K, Wagner D, Kim Y, et al. Sarcopenia predicts costs among patients undergoing major abdominal operations. Surgery 2016; 160: 1162-71.
17. Kirk PS, Friedman JF, Cron DC, Terjimanian MN, Wang SC, Campbell DA, et al. One-year postoperative resource utilization in sarcopenic patients. The Journal of surgical research 2015; 199: 51-5.
18. Sheetz KH, Waits SA, Terjimanian MN, Sullivan J, Campbell DA, Wang SC, et al. Cost of major surgery in the sarcopenic patient. Journal of the American College of Surgeons 2013; 217: 813-8.
19. Gornick ME, Eggers PW, Reilly TW, Mentnech RM, Fitterman LK, Kucken LE, et al. Effects of race and income on mortality and use of services among Medicare beneficiaries. The New England journal of medicine 1996; 335: 791-9.
20. Kroneman M, Boerma W, van den Berg M, Groenewegen P, de Jong J, van Ginneken E. Netherlands: Health System Review. Health systems in transition 2016; 18: 1-240.
21. Obama B. United States Health Care Reform: Progress to Date and Next Steps. JAMA 2016; 316: 525-32.
22. Eurotransplant. [29 December 2016]; Available from: www.eurotransplant.org.
23. Mazzaferro V, Regalia E, Doci R, Andreola S, Pulvirenti A, Bozzetti F, et al. Liver transplantation for the treatment of small hepatocellular carcinomas in patients with cirrhosis. The New England journal of medicine 1996; 334: 693-9.
24. van Vledder MG, Levolger S, Ayez N, Verhoef C, Tran TC, IJzermans JN. Body composition and outcome in patients undergoing resection of colorectal liver metastases. Br J Surg 2012; 99: 550-7.
25. Shen W, Punyanitya M, Wang Z, Gallagher D, St-Onge MP, Albu J, et al. Total body skeletal muscle and adipose tissue volumes: estimation from a single abdominal cross-sectional image. J Appl Physiol (1985) 2004; 97: 2333-8.
26. Martin L, Birdsell L, Macdonald N, Reiman T, Clandinin MT, McCargar LJ, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol 2013; 31: 1539-47.
27. Sinclair M, Poltavskiy E, Dodge JL, Lai JC. Frailty is independently associated with increased hospitalisation days in patients on the liver transplant waitlist. World J Gastroenterol 2017; 23: 899-905.

28. Dasarathy S, Merli M. Sarcopenia from mechanism to diagnosis and treatment in liver disease. J Hepatol 2016; 65: 1232-44.

29. Levolger S, van Vugt JL, de Bruin RW, IJzermans JN. Systematic review of sarcopenia in patients operated on for gastrointestinal and hepatopancreatobiliary malignancies. Br J Surg 2015; 102: 1448-58.

30. Rolland Y, Abellan van Kan G, Gillette-Guyonnet S, Vellas B. Cachexia versus sarcopenia. Current opinion in clinical nutrition and metabolic care 2011; 14: 15-21.

31. Fujiwara N, Nakagawa H, Kudo Y, Tateishi R, Taguri M, Watadani T, et al. Sarcopenia, intramuscular fat deposition, and visceral adiposity independently predict the outcomes of hepatocellular carcinoma. J Hepatol 2015; 63: 131-40.

32. Montano-Loza AJ, Duarte-Rojo A, Meza-Junco J, Baracos VE, Sawyer MB, Pang JX, et al. Inclusion of Sarcopenia Within MELD (MELD-Sarcopenia) and the Prediction of Mortality in Patients With Cirrhosis. Clinical and translational gastroenterology 2015; 6: e102.

33. Yoshizumi T, Shirabe K, Nakagawara H, Ikegami T, Harimoto N, Toshima T, et al. Skeletal muscle area correlates with body surface area in healthy adults. Hepatol Res 2014; 44: 313-8.

34. Meza-Junco J, Montano-Loza AJ, Baracos VE, Prado CM, Bain VG, Beaumont C, et al. Sarcopenia as a prognostic index of nutritional status in concurrent cirrhosis and hepatocellular carcinoma. J Clin Gastroenterol 2013; 47: 861-70.

35. Montano-Loza AJ, Meza-Junco J, Baracos VE, Prado CM, Ma M, Meeberg G, et al. Severe muscle depletion predicts postoperative length of stay but is not associated with survival after liver transplantation. Liver Transpl 2014; 20: 640-8.

36. Dingemans AM, de Vos-Geelen J, Langen R, Schols AM. Phase II drugs that are currently in development for the treatment of cachexia. Expert Opin Investig Drugs 2014; 23: 1655-69.

37. Zhou X, Wang JL, Lu J, Song Y, Kwak KS, Jiao Q, et al. Reversal of cancer cachexia and muscle wasting by ActRIIB antagonism leads to prolonged survival. Cell 2010; 142: 531-43.

38. Carey EJ, Lai JC, Wang CW, Dasarathy S, Lobach I, Montano-Loza AJ, et al. A multicenter study to define sarcopenia in patients with end-stage liver disease. Liver Transpl 2017; 23: 625-33.

39. Harries L, Schrem H, Stahmeyer JT, Krauth C, Amelung VE. High resource utilization in liver transplantation-how strongly differ costs between the care sectors and what are the main cost drivers?: a retrospective study. Transpl Int 2017; 30: 621-37.

SUPPLEMENTARY MATERIAL

Supplementary table 1. Multivariable linear regression analysis for the total hospital costs per patient in a subgroup of patients without hepatocellular carcinoma (n=148).

	B, Euros	Standard Error, Euros	p-value
Skeletal muscle index (cm^2/m^2)	-692	211	0.028
Sex (female)	-1,183	4,890	0.809
Age at moment of listing (years)	-109	207	0.597
MELD score at moment of listing	81	459	0.860
Complications before listing (yes)	3,403	6,054	0.575
Time on the waiting list (days)	29	8	0.001

Abbreviations: MELD, Model for End-stage Liver Disease; HCC, Hepatocellular Carcinoma

CHAPTER 14

Rationale and Study Design of a Randomized Controlled Trial to Reduce Fatigue and Increase Quality of Life with a Rehabilitation Program in Patients with Cancer of the Liver, Pancreas or Biliary Tract Undergoing Surgery

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ABSTRACT

Background: Currently, there is no standardized postoperative guidance and support program for patients who undergo surgery for cancer of the hepatopancreatobiliary (HPB) tract. Nevertheless, the incidence of fatigue complaints is high in these patients. Previous trials have shown beneficial effects of rehabilitation programs in patients with a relatively good prognosis and a high likelihood of reaching a functional status after treatment that is comparable with pre-disease levels compared with HPB cancer patients, who have a poor prognosis.

Objective: We initiate a randomized controlled trial to investigate the effect of a multidimensional rehabilitation program in HPB cancer patients aimed to reduce fatigue complaints and increase quality of life (QoL) after surgery.

Design: In this randomized controlled trial, patients aged ≥18 years with pancreatic, biliary tract, or liver cancer undergoing surgery will be included. Patients will be randomly assigned to an intervention arm (rehabilitation program) or control arm (standard care) in an 1:1 ratio.

Setting: This study will be conducted in three tertiary referral centers in the Netherlands. The intervention will take place in a local setting.

Intervention: Patients in the intervention arm will be offered a rehabilitation program consisting of physical exercise therapy, solution-focused therapy and dietary consultation.

Outcome: The primary outcome is fatigue, assessed using the Multimodal Fatigue Inventory (MFI). Secondary outcomes are QoL, body composition, muscle strength, physical fitness, and overall survival. Finally, cost-effectiveness of the intervention will be assessed.

Discussion: This randomized controlled trial will assess the efficacy of a rehabilitation program to reduce fatigue complaints in a highly complicated patient population with a relatively poor prognosis.

INTRODUCTION

Cancer treatment has improved considerably in recent years. One-year overall survival rates of breast and colon cancer patients are as high as 97% and 80%, respectively [1, 2]. For hepatopancreatobiliary (HPB) cancers, surgical resection is often considered the only curative treatment which is only feasible in up to 25% of patients. One-year overall survival rates of primary hepatic and pancreatic cancer patients in the Netherlands remain as low as 39% and 24% respectively [2]. Besides tumor characteristics, factors such as frailty limit survival in cancer patients [3]. Frailty is a state of increased vulnerability of an individual towards stressors, leading to an increased risk of developing adverse health outcomes [3]. A hallmark sign is fatigue, the most frequently reported symptom in cancer patients during and after treatment [3]. In gastrointestinal cancer patients, almost 30% of patients experience severe fatigue [4, 5]. Low physical activity is independently associated with severe fatigue before cancer treatment [4]. During cancer treatment, fatigue rates in patient populations vary between 25-99% [6]. After successful cancer treatment, 19-38% of disease-free cancer survivors remain fatigued, underlining its persistent character [4, 6]. In our own daily practice, we experience that fatigue in the first months is reported very frequently, particularly after major HPB cancer surgery. Although the exact pathogenesis of fatigue remains unknown, disturbances in physiology, biochemistry and psychology are considered to contribute to this multifactorial symptom [7]. Another key determinant of fatigue may be attributed to age- and disease-related muscle depletion (i.e. sarcopenia) [8]. Its prevalence is common in HPB cancer patients, exceeding rates of 50% in our population [9, 10]. These patients experience muscle weakness that leads to physical impairment, increased fatigue and decreased quality of life [5]. Furthermore, such frail patients are more prone to a reduced therapy effect, complications and increased chemotherapy toxicity [11].

The cancer itself, as well as the sequelae after surgery or chemo(radiation)therapy, may lead to physical and psychosocial impairment [12]. As patients experience increased disease related fatigue, fear/anxiety, depression, muscle mass and strength loss, and subsequently a lower exercise tolerance, they are at great risk of spiraling down a vicious circle which progressively enhances these symptoms and further impairs their quality of life and self-management capacity [13, 14]. Although this series of events is well recognized as a potential course after major HPB surgery, monitoring of patients' well-being after treatment frequently remains a neglected, unstandardized aspect of care. If fatigue complaints and weakness are recognized, it is still unknown how to offer these patients the best supportive care.

Background and rationale

Since multiple dimensions (physical, emotional and cognitive) seem to be involved in fatigue, a multidimensional approach should be opted for [15]. In patients who have been treated for cancer, psychotherapy was shown to significantly reduce fatigue [6]. Particularly interventions with a more general approach, aiming at psychological distress, mood and physical symptoms, are effective in reducing fatigue [16] Furthermore, physical exercise has been demonstrated to reduce fatigue in cancer patients [17, 18] and cancer survivors [19, 20]. The authors of a recent Cochrane review performed a meta-analysis compromising 56 studies (4,068 participants with various cancers) that showed that physical exercise therapy can be regarded as beneficial for patients with cancer-related fatigue during the post-cancer therapy, particularly in patients with solid tumors [17].

Previously, van Weert *et al.* [21] compared the effect of combined physical training and cognitive-behavioral therapy with physical training only and no intervention on cancer-related fatigue in cancer survivors (63.2% breast cancer, 19.7% hematological cancer, 7.9% gynecological cancer, and 9.2% other cancer). Physical training combined with cognitive-behavioral therapy and physical training alone had significant beneficial effects on fatigue compared with no intervention. Physical training was as effective as physical training with cognitive-behavioral therapy. The study population included colorectal and breast cancer patients with relatively good prognosis. Furthermore, the physical exercise therapy program in this study included some elements of coping with fatigue, goal setting and exercise-relaxation balance activities, which may have reduced the effect of the cognitive-behavioral therapy. Solution focused therapy has also been proven effective in various patient populations. A great advantage of solution focused therapy is that a fewer number of sessions is needed to be effective compared with cognitive-behavioral therapy [22, 23]. This translates into lower costs and could possibly lead to greater patient adherence.

These previous studies have been conducted in cancer patients with a relatively good prognosis, such as breast and colorectal cancer [19-21, 24-30]. These populations generally reach a functional status after treatment that is comparable with pre-disease levels and patients may return to regular societal participation. Therefore, the results in these patient populations may not apply to patients undergoing surgical resection of malignancies with a more dismal outcome and high morbidity even mortality in the first years after surgery, such as HPB cancer patients. In addition, HPB cancer patients experience a greatly impaired quality of life according to data from the European Organisation for Research and Treatment of Cancer (EORTC) [31]. The mean global health status or quality of life in HPB cancer patients (all stages) on the EORTC Quality of Life

Questionaire-C30 is 56 points on a scale of 0-100, which is amongst the lowest scores for cancer patients. For example, breast cancer patients, colorectal cancer patients and the general population score 62, 61, and 71.2, respectively [31]. Researchers in- [32, 33] and outside [34, 35] of our research group have demonstrated that quality of life is greatly impaired in HPB cancer patients, including candidates for surgery. In particular in patients who are severely diseased and consequently with worsened survival, the quality of life is lowest [35].

Aim

Although pre- and perioperative care (i.e. the introduction of the ERAS program) have greatly been improved during recent years, no standardized postoperative guidance and support, except a protocol follow-up consultation at the surgical outpatient department, is offered for HPB cancer patients who undergo major surgical resection. Therefore, we initiate a randomized controlled trial (RCT) to investigate the effect of rehabilitation in HPB cancer patients, known for a high incidence of fatigue complaints. The aim of our study is to assess the efficacy of a patient-tailored, supervised 12-week rehabilitation program consisting of physical therapy and solution focused psychotherapy in a well-defined population of surgically treated HPB cancer patients with relatively poor prognosis.

Population

All consecutive patients undergoing surgery for HPB malignancies (i.e. bile duct cancer, pancreatic cancer, hepatic cancer) in Erasmus MC University Medical Center (Rotterdam, the Netherlands), Academic Medical Center (Amsterdam, the Netherlands) and Leiden University Medical Center (Leiden, the Netherlands) will be considered for eligibility for study participation. Particularly these patients may benefit from the described intervention. After all, they have a dismal prognosis, but are considered fit enough to undergo surgery as this is the only treatment option with curative intent.

Inclusion criteria

In order to be eligible to participate in this study, a subject must meet all of the following criteria:

- Undergoing curative intent surgery for HPB malignancies;
- Clinically suspect or histologically confirmed liver, bile duct or pancreatic carcinoma;
- Life expectancy of at least 6 months;
- Mild to severe fatigue after hospital discharge, according to the MFI;
- Able to read and understand the Dutch language;
- Written informed consent.

Exclusion criteria

A potential subject who meets any of the following criteria will be excluded from participation in this study:

- Treatment with adjuvant chemo(radiation)therapy
- Bone metastases or other high risk of fractures;
- Not able to perform basic activities of daily living (ECOG ≥3);
- Decompensated heart disease, uncontrolled hypertension (systolic blood pressure > 200 mmHg or diastolic blood pressure > 110 mmHg), heart failure (NYHA Class II or greater) or chronic obstructive pulmonary disease causing fatigue;
- Living in nursing homes;
- Cognitive impairment;
- BMI <15 kg^2/m^2, >5% weight loss per month or other health problems that would not allow physical exercise training;
- Anxiety or depression requiring psychiatric consultation;
- Cancer treatment in the previous 5 years (except basal skin cancer);
- Participation in other studies containing elements of physical exercise or psychological therapy.

STUDY OUTCOMES

The main outcome is general fatigue. Symptoms of fatigue are assessed using the Multidimensional Fatigue Inventory (MFI) [15]. This 20-item questionnaire includes the following five scales: general fatigue, physical fatigue, reduced activity, reduced motivation, and mental fatigue. Each question can be scored with 1 to 5 points, with sub scores for the five dimensions ranging from 4 to 20. Higher scores indicate increased fatigue. Fatigue will be assessed preoperatively, postoperatively (before the rehabilitation program), directly after the rehabilitation program, and 6 and 12 months after surgery. Besides quality of life, other secondary outcome measures are: body composition (i.e. skeletal muscle mass and density, visceral and subcutaneous adipose tissue mass), muscle strength, frailty, anxiety and depression, and physical fitness. Measurements will be repeated at 6 and 12 months after surgery to explore long-term effects. We hypothesize that offering a combined physical exercise and psychotherapy program halts the vicious circle with down spiraling character. We assume that the combined treatment as proposed here is the best method to approach a multidimensional and multifactorial symptom such as fatigue, which will work synergistically and show the greatest result. Nevertheless, the individual effects of physical exercise therapy and psychotherapy remain unknown. Therefore, we also measure body composition and

muscle function and –strength to investigate if there are any differences in fatigue and quality of life between patients with lowmuscle mass, who are wasting muscle mass, or (re)gaining muscle mass. These data could be used to further guide future studies. Moreover, the effect on overall survival will be assessed. Finally, a cost-effectiveness analysis will be performed.

Quality of life

The (health-related) quality of life will be assessed using the EORTC-Quality of Life-C30 questionnaire (version 3) and Short Form 36. Both Dutch versions of these questionnaires have been validated [36, 37].

Cardiopulmonary fitness

One of the most relevant characteristics of physical fitness is cardiopulmonary fitness. The golden standard to measure cardiopulmonary fitness is a cardiopulmonary exercise test (CPET) [38, 39]. With a CPET, the entire cardiopulmonary system is tested. The oxygen use, carbon dioxide production, breathing patterns are measured and an electrocardiography is performed during a bike test. The CPETs will be performed at baseline to tailor a physical exercise program for each patient.

Body composition measurements

All patients will undergo computed tomography (CT) examinations preoperatively (within 4 weeks before surgery), at baseline (start rehabilitation program) and after 6 and 12 months hereafter. All examinations, except those after 6 months, are part of standard care. The cross-sectional skeletal muscle area (cm^2) will be measured on the level of the third lumbar vertebra (L3). This area will be adjusted for patients' height, resulting in the L3 muscle index (cm^2/m^2). This measure is strongly correlated with total body skeletal muscle mass and is known for its high reliability and great reproducibility [10, 40]. Additionally, the amount of visceral adipose tissue will be measured [41].

Muscle strength

Muscle strength is a predictor for survival in patients of average and higher age. Furthermore, muscle strength of the upper extremities is an important characteristic of functional decline in elderly [42]. Muscle weakness results in decreased self-management and a higher rate of dependency. The strength of the quadriceps muscle will be measured using a handheld dynamometer. The peak force during contraction will be measured for 5 consecutive seconds [42]. A JAMAR® hand dynamometer will be used to measure muscle strength of the upper extremities, according to a previously described method [43]; sitting on a chair with the shoulders adducted and neutrally rotated, the arm

in vertical (next to the body) position and the wrist in neutral position, the subject is instructed to squeeze the grip with maximum strength. Decreased strength is defined as <85% of the population-based value, according to Webb and colleagues [44]. These sex- and age specific cut-off values are routinely being used in physiotherapist practices in the Netherlands.

General physical performance

The performance status of patients during daily activities will be investigated using the Karnofsky performance scale. This is a scale ranging from 0 (dead) until 100 (normal general performance without complaints). An in- or decrease of 10 points will be used to detect significant differences. Furthermore, the timed sit-up-and-go test [45] as well as the six-minute walk test [46] will be performed.

Frailty and anxiety and depression assessment

Frailty will be assessed using the Groningen Frailty Indicator (GFI) [47]. The GFI has been developed as a simple screening instrument for frailty. It screens on physical, cognitive, social, and emotional items. The maximum score is 15 points. Patients scoring 5 or more points are considered frail. Anxiety and depression mood will be measured using the Hospital Anxiety and Depression Scale (HADS), consisting of the anxiety (HADS-A) and depression (HADS-D) subscales. Both subscales have a score ranging from 0 to 21 with higher scores indicating more anxiety and depression [48].

Body height and weight

Body height and weight will be measured on the described time points and body mass index (BMI) will be calculated.

Overall survival

The possible effect of the intervention on overall survival will be assessed on various time points (1 and 3-year) after the intervention. Survival status will be subtracted from the municipal registration system or the electronic patient file.

Randomisation, blinding and treatment allocation

Patients will be included on the preoperative surgical outpatient department and with hospital discharge patients will randomly be assigned to the treatment (rehabilitation program) or control (standard care) group in a 1:1 ratio. Randomization will be conducted by an independent researcher using a computer-generated randomization procedure with variable block length. Neither patients nor therapists will be blinded for

the treatment group. Stratification for gender and age will be conducted. All therapists will be blinded for the Multidimensional Fatigue Inventory (MFI) score that will be obtained before the start of the rehabilitation program.

Study procedures

The intervention arm will be offered close to the patients' home. Patients in the control arm are offered standard medical care according to the local protocols.

Physical exercise therapy

The physical exercise therapy program will start four weeks after hospital discharge for a duration of 12 weeks. The intervention group is offered a supervised and personalized exercise program that is aimed at both cardiorespiratory fitness (aerobic training) and muscle strength (resistance training) [49], which is also considered feasible in patients receiving chemotherapy [30]. The program is developed in a close collaboration between physiotherapists, exercise physiologists, revalidation specialists, and medical and surgical oncologists. For each patient, an intake or baseline exercise measurement (i.e. CPET and muscle function tests) will be performed. Subsequently, the program will be specified to each patient's personal preferences and physical fitness. Hereafter, patients will train for a period of one hour twice a week under the supervision of a physical therapist. Patients are offered the following program: muscle strength exercises for 30 minutes per training session, starting with 12 repetitions at 70% of the one repetition maximum (1RM) and end with two series at 80% 1RM. One training exists of a minimum of six exercises: 1. Vertical row (longissimus, biceps brachii, rhomboideus); 2. Leg press (quadriceps, glutei, gastrocnemicus); 3. Bench press (pectoralis major, triceps); 4. Pull over (pectoralis, triceps brachii, deltoideus, trapezius); 5. Abdominal crunch (rectus abdominis); 6. Lunge (quadriceps, glutei, hamstrings). The aerobic training will be performed for the following 30 minutes. This program may be adjusted according to the individual patient's preferences and abilities. We aim to increase or decrease the intensity of the training according to the Borg scale of perceived exertion, resulting in an exercise intensity with a Borg scale of around 14. The total workload will be 50-80% of the maximal workload. The heart rate should be between 60-90% of the maximum heart rate, which can be estimated as 220 minus age and is assessed during the baseline CPET [30]. Patients will be informed about and motivated to perform daily exercise according to the Dutch Consensus on Healthy Exercise [50].

Solution focused (psycho)therapy

Solution focused therapy has empirically been validated and shown successful in patients with chronic diseases [51]. It has improved symptoms of fatigue in patient populations with somatic diseases, such as patients with inflammatory bowel disease, by our research group [22, 23]. It was considered more feasible with fewer drop outs than problem solved therapy [23]. Contrary to other forms of psychotherapy, solution focused therapy is goal-oriented, future-focused and focuses on solutions to patients' problems rather than on their problems. This approach assumes that everyone has some knowledge of what would make their life better, as well as some necessary coping skills. It is a short-term psychological intervention during which patients will be offered various interventions to channel their attention towards constructing solutions, including: goal setting, compliments, miracle questions, scaling questions, coping questions, and exception questions. The solution focused therapy will be offered in six sessions of one hour during twelve weeks and will be focused on fatigue management.

Dietary consultation

A specialized dietician offers patients in the intervention arm postoperative dietary consultation with nutritional assessment. The individual energy and protein need is assessed using the Harris-Benedict method. Using this method, the basic metabolism will be calculated with the number of calories needed adjusted for the rate of activity. If needed, additional consultations can be scheduled. Patients in the control arm receive standard dietary care; only in the postoperative phase during hospital admission, whereas the intervention arm will be followed after discharge.

Withdrawal and replacement of individual subjects

Subjects can leave the study at any time for any reason if they wish to do so without any consequences. The investigator can decide to withdraw a subject from the study for urgent medical reasons. Withdrawn subjects will not be replaced. Overall- and disease free survival will be assessed.

Premature termination of the study

Although we have currently no reason to believe that the intervention is too intensive for the patients to be included, the study will prematurely be terminated when the intervention seems to be unfeasible.

If the drop out of included patients exceeds the estimated dropout rate and inclusion of patients within the given study period will not be possible, the study will prematurely be terminated.

STATISTICAL ANALYSIS

Statistical analyses will be performed using SPSS statistical software. Two-sided p-values <0.05 will be considered significant. Adjusted analysis with prognostic baseline characteristics (e.g., gender, age, disease stage, radicality of resection) will be performed. This results in increased statistical power, corrects for baseline differences (based on chance) and provides better individualized treatment effect estimates [52]. Analyses of variance (ANOVA) and longitudinal data analyses will be used to compare fatigue and quality of life scores between the two groups, as well as measures of skeletal muscle mass and function, depression and anxiety, frailty, and functional capacity. Furthermore, differences in outcome between baseline and the sequential measurements will be performed. Clinically important changes will be estimated as effect sizes using Cohen's guidelines. An effect size of 0.2 will be considered as small, 0.5 as medium, and 0.8 as large.

Sample size

General fatigue (assessed with the Multidimensional Fatigue Inventory (MFI)) is the primary outcome. To detect a medium difference in MFI-General fatigue (Cohen's d = 0.50) with a power of 0.80 and alpha of 0.05 (two-sided), both groups should include 64 patients. With an expected 20% drop out rate [24], a total of 154 patients are needed. Approximately 120 HCC, 150 pancreatic cancer and 30 bile duct cancer patients undergo surgery in the three participating centers annually. Consequently, patient inclusion should be finished within something more than one year considering an inclusion rate of 50%. Follow-up will be conducted until death to explore a possible association between the interventional and survival benefit.

Cost-effectiveness analysis

A cost-utility analysis will be performed to identify the most cost-effective treatment (i.e. standard care or rehabilitation program), according to the Dutch guidelines. Both societal and healthcare provider perspectives will be addressed. The time horizon will be 12 months from surgery. The quality of life and costs will be assessed for each study arm. QALYs (based on the EQ-5D-5L) will be the outcome measure for quality of life and costs per QALY for the effectiveness analysis. If no differences in QALYs between the treatment arms are found, a cost-effectiveness analysis will be performed from both societal and healthcare provider perspectives, based on the QLQ-C30. The incremental cost-effectiveness ratios (ICERs), defined as differences in costs of the rehabilitation arm versus the standard care arm divided by the average change in QALYs or HRQoL, of the arms will be calculated. Sensitivity analyses will be performed to test the sensitivity of

various costs per unit of resource. No discounting for costs and effects will be used, due to the relatively short time horizon of 12 months. Direct and indirect costs will be accounted for in the cost analysis and estimated by multiplying resource utilization with the cost per unit of resource following the micro-costing method which is based on comprehensive 'bottom-up' analyses and included costs of employment, material and equipment. In other words, direct costs of each treatment arm will be accounted for. Hospital databases will be used to collect costs. Direct non-medical costs (e.g., travel costs of patients) will be determined using ZIP (postal) codes. The SF-HLW and friction cost approach will be used to take the indirect non-medical costs into account. Sensitivity analyses will be performed for uncertain and variable input variables to perform robust calculations.

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REFERENCES

1. IKNL. Cijfers over kanker. Available from: http://www.cijfersoverkanker.nl/.
2. Berrino F, Verdecchia A, Lutz JM, Lombardo C, Micheli A, Capocaccia R, et al. Comparative cancer survival information in Europe. Eur J Cancer 2009; 45: 901-8.
3. Fried LP, Hadley EC, Walston JD, Newman AB, Guralnik JM, Studenski S, et al. From bedside to bench: research agenda for frailty. Science of aging knowledge environment : SAGE KE 2005; 2005: pe24.
4. Goedendorp MM, Gielissen MF, Verhagen CA, Peters ME, Bleijenberg G. Severe fatigue and related factors in cancer patients before the initiation of treatment. Br J Cancer 2008; 99: 1408-14.
5. Rizzoli R, Reginster JY, Arnal JF, Bautmans I, Beaudart C, Bischoff-Ferrari H, et al. Quality of life in sarcopenia and frailty. Calcif Tissue Int 2013; 93: 101-20.
6. Gielissen MF, Verhagen S, Witjes F, Bleijenberg G. Effects of cognitive behavior therapy in severely fatigued disease-free cancer patients compared with patients waiting for cognitive behavior therapy: a randomized controlled trial. J Clin Oncol 2006; 24: 4882-7.
7. Ryan JL, Carroll JK, Ryan EP, Mustian KM, Fiscella K, Morrow GR. Mechanisms of cancer-related fatigue. Oncologist 2007; 12 Suppl 1: 22-34.
8. Rolland Y, Abellan van Kan G, Gillette-Guyonnet S, Vellas B. Cachexia versus sarcopenia. Current opinion in clinical nutrition and metabolic care 2011; 14: 15-21.
9. Levolger S, van Vledder MG, Muslem R, Koek M, Niessen WJ, de Man RA, et al. Sarcopenia impairs survival in patients with potentially curable hepatocellular carcinoma. J Surg Oncol 2015; 112: 208-13.
10. Levolger S, van Vugt JL, de Bruin RW, JN IJ. Systematic review of sarcopenia in patients operated on for gastrointestinal and hepatopancreatobiliary malignancies. Br J Surg 2015; [Epub ahead of print].
11. Prado CM, Birdsell LA, Baracos VE. The emerging role of computerized tomography in assessing cancer cachexia. Curr Opin Support Palliat Care 2009; 3: 269-75.
12. Hamaker ME, Prins MC, Schiphorst AH, van Tuyl SA, Pronk A, van den Bos F. Long-term changes in physical capacity after colorectal cancer treatment. J Geriatr Oncol 2014.
13. Curt GA. Impact of fatigue on quality of life in oncology patients. Semin Hematol 2000; 37: 14-7.
14. Lucia A, Earnest C, Perez M. Cancer-related fatigue: can exercise physiology assist oncologists? Lancet Oncol 2003; 4: 616-25.
15. Smets EM, Garssen B, Bonke B, De Haes JC. The Multidimensional Fatigue Inventory (MFI) psychometric qualities of an instrument to assess fatigue. J Psychosom Res 1995; 39: 315-25.

16. Goedendorp MM, Gielissen MF, Verhagen CA, Bleijenberg G. Psychosocial interventions for reducing fatigue during cancer treatment in adults. Cochrane Database Syst Rev 2009: CD006953.

17. Cramp F, Byron-Daniel J. Exercise for the management of cancer-related fatigue in adults. Cochrane Database Syst Rev 2012; 11: CD006145.

18. Mishra SI, Scherer RW, Snyder C, Geigle PM, Berlanstein DR, Topaloglu O. Exercise interventions on health-related quality of life for people with cancer during active treatment. The Cochrane database of systematic reviews 2012; 8: CD008465.

19. Korstjens I, May AM, van Weert E, Mesters I, Tan F, Ros WJ, et al. Quality of life after self-management cancer rehabilitation: a randomized controlled trial comparing physical and cognitive-behavioral training versus physical training. Psychosom Med 2008; 70: 422-9.

20. May AM, Korstjens I, van Weert E, van den Borne B, Hoekstra-Weebers JE, van der Schans CP, et al. Long-term effects on cancer survivors' quality of life of physical training versus physical training combined with cognitive-behavioral therapy: results from a randomized trial. Support Care Cancer 2009; 17: 653-63.

21. van Weert E, May AM, Korstjens I, Post WJ, van der Schans CP, van den Borne B, et al. Cancer-related fatigue and rehabilitation: a randomized controlled multicenter trial comparing physical training combined with cognitive-behavioral therapy with physical training only and with no intervention. Phys Ther 2010; 90: 1413-25.

22. Vogelaar L, van't Spijker A, Timman R, van Tilburg AJ, Bac D, Vogelaar T, et al. Fatigue management in patients with IBD: a randomised controlled trial. Gut 2014; 63: 911-8.

23. Vogelaar L, Van't Spijker A, Vogelaar T, van Busschbach JJ, Visser MS, Kuipers EJ, et al. Solution focused therapy: a promising new tool in the management of fatigue in Crohn's disease patients psychological interventions for the management of fatigue in Crohn's disease. J Crohns Colitis 2011; 5: 585-91.

24. Adamsen L, Quist M, Andersen C, Moller T, Herrstedt J, Kronborg D, et al. Effect of a multimodal high intensity exercise intervention in cancer patients undergoing chemotherapy: randomised controlled trial. Bmj 2009; 339: b3410.

25. Kampshoff CS, Buffart LM, Schep G, van Mechelen W, Brug J, Chinapaw MJ. Design of the Resistance and Endurance exercise After ChemoTherapy (REACT) study: a randomized controlled trial to evaluate the effectiveness and cost-effectiveness of exercise interventions after chemotherapy on physical fitness and fatigue. BMC Cancer 2010; 10: 658.

26. Kampshoff CS, Chinapaw MJ, Brug J, Twisk JW, Schep G, Nijziel MR, et al. Randomized controlled trial of the effects of high intensity and low-to-moderate intensity exercise on physical fitness and fatigue in cancer survivors: results of the Resistance and Endurance exercise After ChemoTherapy (REACT) study. BMC Med 2015; 13: 275.

27. Sancho A, Carrera S, Arietaleanizbeascoa M, Arce V, Gallastegui NM, Gine March A, et al. Supervised physical exercise to improve the quality of life of cancer patients: the EFICANCER randomised controlled trial. BMC Cancer 2015; 15: 40.

28. Travier N, Velthuis MJ, Steins Bisschop CN, van den Buijs B, Monninkhof EM, Backx F, et al. Effects of an 18-week exercise programme started early during breast cancer treatment: a randomised controlled trial. BMC Med 2015; 13: 121.

29. van Waart H, Stuiver MM, van Harten WH, Geleijn E, Kieffer JM, Buffart LM, et al. Effect of Low-Intensity Physical Activity and Moderate- to High-Intensity Physical Exercise During Adjuvant Chemotherapy on Physical Fitness, Fatigue, and Chemotherapy Completion Rates: Results of the PACES Randomized Clinical Trial. J Clin Oncol 2015; 33: 1918-27.

30. van Waart H, Stuiver MM, van Harten WH, Sonke GS, Aaronson NK. Design of the Physical exercise during Adjuvant Chemotherapy Effectiveness Study (PACES): a randomized controlled trial to evaluate effectiveness and cost-effectiveness of physical exercise in improving physical fitness and reducing fatigue. BMC Cancer 2010; 10: 673.

31. Scott NW, Fayers PW, Aaronson NL, Bottomley A, de Graeff A, Groenvold M, et al. EORTC QLQ-C30 Reference Values. 2008; Available from: http://groups.eortc.be/qol/sites/default/files/img/newsletter/reference_values_manual2008.pdf.

32. Uitdehaag MJ, Verschuur EM, van Eijck CH, van der Gaast A, van der Rijt CC, de Man RA, et al. Problems and needs in patients with incurable esophageal and pancreaticobiliary cancer: a descriptive study. Gastroenterol Nurs 2015; 38: 42-54.

33. Morak MJ, Pek CJ, Kompanje EJ, Hop WC, Kazemier G, van Eijck CH. Quality of life after adjuvant intra-arterial chemotherapy and radiotherapy versus surgery alone in resectable pancreatic and periampullary cancer: a prospective randomized controlled study. Cancer 2010; 116: 830-6.

34. Huang G, Chen X, Lau WY, Shen F, Wang RY, Yuan SX, et al. Quality of life after surgical resection compared with radiofrequency ablation for small hepatocellular carcinomas. Br J Surg 2014; 101: 1006-15.

35. Meier A, Yopp A, Mok H, Kandunoori P, Tiro J, Singal AG. Role functioning is associated with survival in patients with hepatocellular carcinoma. Qual Life Res 2015; 24: 1669-75.

36. Aaronson NK, Ahmedzai S, Bergman B, Bullinger M, Cull A, Duez NJ, et al. The European Organization for Research and Treatment of Cancer QLQ-C30: a quality-of-life instrument for use in international clinical trials in oncology. J Natl Cancer Inst 1993; 85: 365-76.

37. Aaronson NK, Muller M, Cohen PD, Essink-Bot ML, Fekkes M, Sanderman R, et al. Translation, validation, and norming of the Dutch language version of the SF-36 Health Survey in community and chronic disease populations. J Clin Epidemiol 1998; 51: 1055-68.

38. Ross RM. ATS/ACCP statement on cardiopulmonary exercise testing. Am J Respir Crit Care Med 2003; 167: 1451; author reply

39. Slaman J, Dallmeijer A, Stam H, Russchen H, Roebroeck M, van den Berg-Emons R, et al. The six-minute walk test cannot predict peak cardiopulmonary fitness in ambulatory adolescents and young adults with cerebral palsy. Arch Phys Med Rehabil 2013; 94: 2227-33.

40. van Vugt JL, Levolger S, Gharbharan A, Koek M, Niessen WJ, Burger JW, et al. A comparative study of software programmes for cross-sectional skeletal muscle and adipose tissue measurements on abdominal computed tomography scans of rectal cancer patients. J Cachexia Sarcopenia Muscle 2016.

41. Cakir H, Heus C, van der Ploeg TJ, Houdijk AP. Visceral obesity determined by CT scan and outcomes after colorectal surgery; a systematic review and meta-analysis. Int J Colorectal Dis 2015; 30: 875-82.

42. Martin HJ, Yule V, Syddall HE, Dennison EM, Cooper C, Aihie Sayer A. Is hand-held dynamometry useful for the measurement of quadriceps strength in older people? A comparison with the gold standard Bodex dynamometry. Gerontology 2006; 52: 154-9.

43. Sultan P, Hamilton MA, Ackland GL. Preoperative muscle weakness as defined by handgrip strength and postoperative outcomes: a systematic review. BMC Anesthesiol 2012; 12: 1.

44. Webb AR, Newman LA, Taylor M, Keogh JB. Hand grip dynamometry as a predictor of postoperative complications reappraisal using age standardized grip strengths. JPEN J Parenter Enteral Nutr 1989; 13: 30-3.

45. Mathias S, Nayak US, Isaacs B. Balance in elderly patients: the "get-up and go" test. Arch Phys Med Rehabil 1986; 67: 387-9.

46. Lord SR, Menz HB. Physiologic, psychologic, and health predictors of 6-minute walk performance in older people. Arch Phys Med Rehabil 2002; 83: 907-11.

47. Schuurmans H, Steverink N, Lindenberg S, Frieswijk N, Slaets JP. Old or frail: what tells us more? J Gerontol A Biol Sci Med Sci 2004; 59: M962-5.

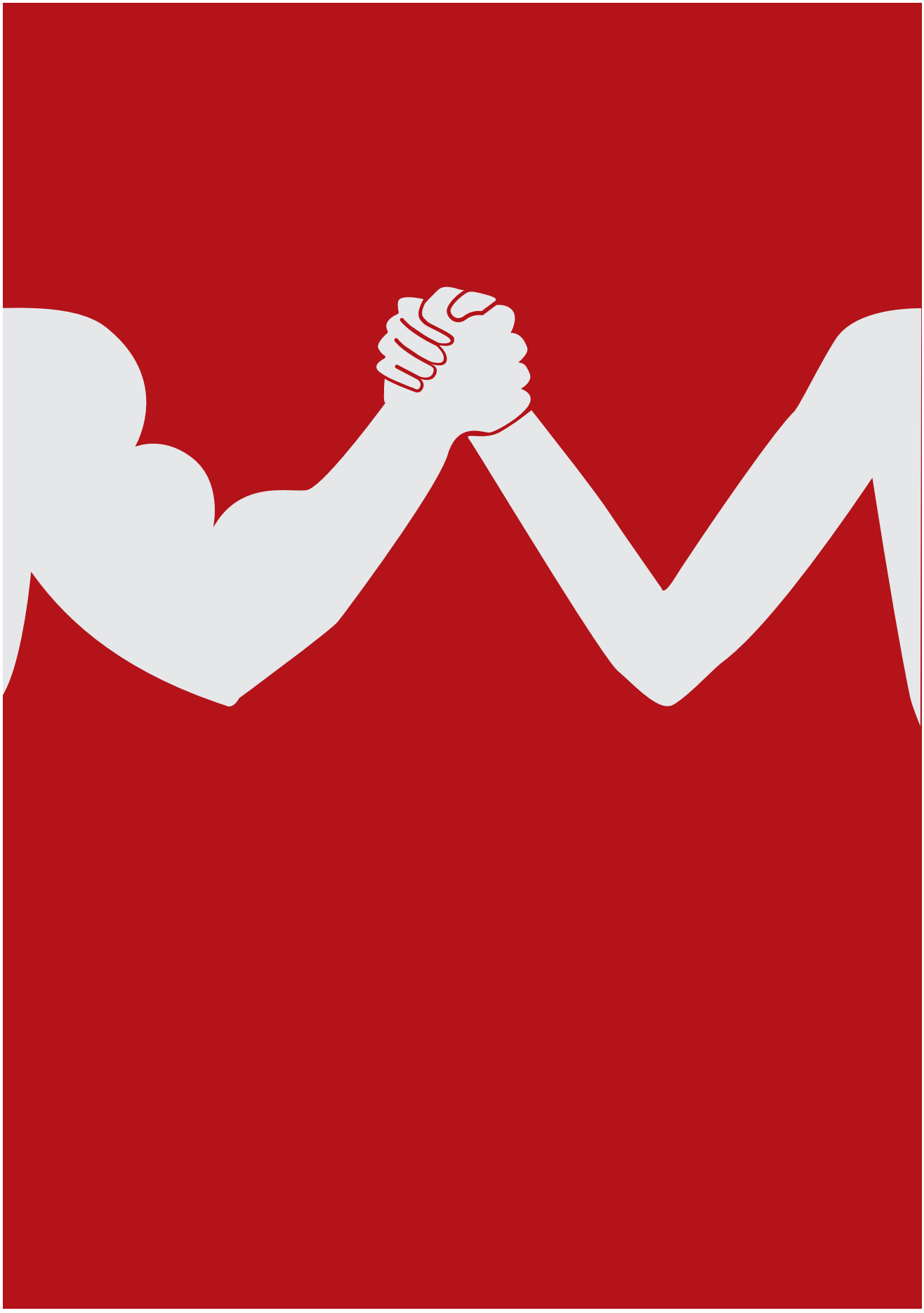
48. Zigmond AS, Snaith RP. The hospital anxiety and depression scale. Acta Psychiatr Scand 1983; 67: 361-70.

49. Courneya KS, Segal RJ, Mackey JR, Gelmon K, Reid RD, Friedenreich CM, et al. Effects of aerobic and resistance exercise in breast cancer patients receiving adjuvant chemotherapy: a multicenter randomized controlled trial. J Clin Oncol 2007; 25: 4396-404.

50. Kemper HG OW, Stiggelbout M. . Consensus over de Nederlandse Norm voor Gezond Bewegen. Tijdschr Sociale Gezondheidszorg 2000; 78: 180-3.

51. Bond C, Woods K, Humphrey N, Symes W, Green L. Practitioner Review: The effectiveness of solution focused brief therapy with children and families: a systematic and critical evaluation of the literature from 1990-2010. J Child Psychol Psychiatry 2013; 54: 707-23.

52. Roozenbeek B, Maas AI, Lingsma HF, Butcher I, Lu J, Marmarou A, et al. Baseline characteristics and statistical power in randomized controlled trials: selection, prognostic targeting, or covariate adjustment? Crit Care Med 2009; 37: 2683-90.



PART VI

THESIS OVERVIEW

VI

CHAPTER 15

Summary and General Discussion
Future Perspectives
Nederlandse Samenvatting

15

SUMMARY AND GENERAL DISCUSSION

In this thesis, we investigated the use of computed tomography (CT) to measure skeletal muscle mass and explored the impact and consequences of low skeletal muscle mass in surgical populations. Skeletal muscle mass may be an objective measure, reflecting a patient's physiological status and reserves accurately. This may particularly be relevant in an era of an increasingly aging population in which cancer is the leading cause of death and liver transplantation is hampered by a shortage of donors. We investigated the long and short-term effects of low skeletal muscle mass in surgical patients, which are described in this thesis.

In **part I** of this thesis we investigated the accuracy of techniques to measure skeletal muscle mass and density on CT. Over the last years, numerous studies have used CT to quantify skeletal muscle mass, for example in medical [1-5] and surgical oncology [6], vascular surgery [7], and transplantation surgery [8, 9] patients. Furthermore, multiple studies measured visceral and/or subcutaneous adipose tissue on CT [10-13]. However, different software programs have been used to perform these body composition analyses, such as FatSeg [13], OsiriX [14], ImageJ [3], and SliceOmatic [2]. To adequately interpret and compare the study results using these different programs, we investigated their comparability in **chapter 2**. We showed excellent inter-software agreement between these four software packages for skeletal muscle, subcutaneous adipose tissue and visceral adipose tissue measurements. Although we found small but statistically significant differences within and between observers in skeletal muscle mass measurements, these differences were found not clinically relevant. Cohen's κ was excellent for the classification of patients with or without sarcopenia. Differences within and between observers were greatest for visceral adipose tissue measurements. After all, the distribution of the intra-abdominal organs (e.g. bowel) can greatly differ between slices. Although previous studies frequently described single slice visceral adipose tissue measurements [10, 13, 15], these measurements may not be clinically applicable and should be reserved for clinical research of patient cohorts rather than individual patients.

Although many research groups consider CT the gold standard for body composition measurements [1, 16], there currently is no standardized method or protocol to perform these measurements. One of the unknown aspects, for example, was the effect of contrast-enhancement of CT examinations. Therefore, we investigated the effect of contrast-enhancement (i.e. unenhanced, portal-venous, and arterial phase) on skeletal muscle mass and density measurements on CT in **chapter 3**. We found statistically significant differences in skeletal muscle mass between contrast-enhancement phases. However, these differences were very small and likely not clinically relevant. Significant

differences in skeletal muscle density were found between the unenhanced and both contrast-enhanced phases, but not between the portal-venous and arterial phase. Similar results were recently described by Van der Werf *et al.* [17]. Based on these results, we recommend using the portal-venous phase, which is routinely performed in cancer and liver transplant patients, to measure skeletal muscle mass and density. Studies using the same contrast-enhancement phase to measure skeletal muscle mass will lead to higher comparability and uniformity of results. International consensus on using the portal venous phase should be advocated.

Despite a great number of studies in which only the psoas muscle was measured [6, 7, 9], we chose to measure the cross-section skeletal muscle area at the level of the third lumbar vertebra (L3), including the rectus abdominis, obliquus internus abdominis, obliquus externus abdominis, transversus abdominis, psoas major, quadratus lumborum and the erector spinae muscles. We support the concept presented by Baracos, who described the psoas muscle as a proxy for sarcopenia as a 'flawed premise' [18]. The psoas muscle alone is not representative for total body skeletal muscle mass for varying reasons: it only includes a low proportion of the total (<10%), comes with higher measurement errors, has a weak correlation with the total lumbar muscle area, and is subject to a high likelihood of isolated atrophy (e.g. due to osteoarthritis of the hip) [18]. In a group of ovarian cancer patients, Rutten *et al.* described a weak correlation between the cross-sectional skeletal muscle area and the bilateral psoas area, both measured using software and calculated using the psoas width and length [19]. Consequently, expert groups advise not to measure one muscle as a sentinel for total skeletal muscle mass [18]. Although psoas measurements could be somewhat less time consuming than cross-sectional skeletal muscle area measurements, the latter method takes a few minutes only [20]. Furthermore, automated software is currently being developed [20, 21], which may facilitate the clinical applicability.

There is still much debate regarding adequate cut-off points for the skeletal muscle index to define sarcopenia. Prado *et al.* used cut-off values based on risk stratification[2]. These have been used commonly by others, yet some studies use different cut-off values, such as the sex-specific median or lowest quartile for skeletal muscle mass [13, 22, 23]. No international consensus has been reached yet and there is insufficient knowledge on skeletal muscle mass in healthy persons.

In **chapter 4** we measured skeletal muscle mass and density on CT in over 1,000 living kidney donors, who may be considered as healthy subjects. We found that skeletal muscle mass and density were significantly associated with sex, age, and body mass

index (BMI). We developed a nomogram and online calculator to estimate a patient's healthy skeletal muscle mass and density, adjusted for these parameters. The nomogram requires external validation.

The consequences of low skeletal muscle mass and density for surgical oncology patients are described in **part II** of this thesis. In **chapter 5** we performed a systematic review on the impact of sarcopenia on outcome after surgery for gastrointestinal and hepatopancreatobiliary cancer surgery. In total, 13 studies with a total of 2,884 patients were included. Overall, we found that sarcopenia was independently associated with reduced overall survival in seven out of ten studies. An association between sarcopenia, postoperative complications, and mortality was observed in patients undergoing partial colectomy or hepatectomy for colorectal cancer in particular. A great variety in methods and definitions of sarcopenia precluded performing meta-analyses, underscoring the need for more uniformity.

In **chapter 6** we investigated the association of functional compromise with postoperative complications and mortality in a cohort of 310 patients undergoing colorectal cancer surgery. Functional compromise was defined as the sarcopenia measured on CT, frailty, and nutritional depletion, both measured using questionnaires. Mortality was significantly higher in patients with sarcopenia compared with patients without sarcopenia (8.8% versus 0.7%, p=0.001; OR 15.5, 95% confidence interval [CI] 2.00-120). Sarcopenia was not predictive for anastomotic leakage or sepsis. The combination of sarcopenia, frailty, and nutritional depletion, however, was strongly predictive for sepsis (OR 25.1, 95% CI 5.11-123, p=0.001), with a sensitivity of 46% and specificity of 97%. An association between low skeletal muscle mass and infectious complications has been described before in multiple studies [24-26]. An explanation of the higher occurrence of infections in patients with low skeletal muscle mass may be an increased systemic inflammatory state [27-29].

In the next two chapters we validated these findings in colorectal cancer patients in two cohorts of patients. First, in **chapter 7**, we found an independent association between an incremental increase in skeletal muscle mass (L3 index, cm^2/m^2) protecting against the occurrence of severe complications (i.e. Clavien Dindo grade ≥3a) in patients undergoing cytoreductive surgery (CRS) with hyperthermic intraperitoneal chemotherapy (HIPEC) for colorectal cancer that has been metastasized to the peritoneum (OR 0.93, 95% CI 0.87-0.99, p=0.018). Second, in **chapter 8** we described the association between skeletal muscle mass and density and both short-term and long-term outcome in patients undergoing surgery for stage I-III colorectal cancer for which we used a prospective, multicenter cohort of 816 patients. As for the studies described

in chapter 6 and chapter 7, we found a significantly higher rate of severe complications in patients with low skeletal muscle mass or density. Low skeletal muscle density was independently associated with the occurrence of severe postoperative complications (OR 1.89, 95% CI 1.11-3.23, p=0.020). Furthermore, we found a higher mortality rate in patients with low skeletal muscle density (3.4% versus 1.0%, p=0.038) and low skeletal muscle mass (3.6% versus 1.7%, p=0.091). In univariable analysis, both low skeletal muscle density and mass were associated with impaired overall survival. The fact that low skeletal muscle mass and density are associated with short-term, but not long-term, outcome suggests an increased vulnerability of colorectal cancer patients towards stressors, such as surgery (i.e. frailty) [30]. Once patients have survived the postoperative period, the effects of low skeletal muscle mass and density disappear. Other risk factors, such as age, tumor stage, and comorbidity, become more important for the clinical course of a given patient. We hypothesize that patients with stage I-III colorectal cancer surviving the perioperative period, generally reach a functional status that is close to pre-disease levels [31]. However, patients undergoing chemotherapy and elderly are at increased risk for functional status decline after cancer treatment [32].

Previous studies described sarcopenia as an independent prognostic factor in patients with perihilar cholangiocarcinoma (PHC) following hepatectomy [22] and as a risk factor for postoperative complications [22, 33]. Unfortunately, only a minority of patients presents with resectable disease. We aimed to investigate the association between low skeletal muscle mass and density in all patients with PHC, regardless of subsequent treatment, in **chapter 9**. Although we did not find an association between low skeletal muscle mass and overall survival, patients with low skeletal muscle density showed a significantly shorter overall survival compared with patients without low skeletal muscle density: 7.0 versus 12.1 months (p=0.004). This effect was time-dependent; an independent association was found within the first six months after presentation (HR 1.78, 95% CI 1.03-3.07, p=0.04), but not after six months (HR 0.68, 95% CI 0.44-1.07, p=0.093) after adjustment for age, tumor size, and suspicion of distant metastases. In conclusion, low skeletal muscle density identified patients with PHC most prone for early death.

Remarkably, we found a long-term effect of sarcopenia in patients with PHC, but not in patients with colorectal carcinoma, in which we found independent short-term effects (i.e. increased postoperative complication and mortality rate) and only long-term effects in univariable analysis. One could hypothesize that once patients have survived the postoperative period, the effects of low skeletal muscle mass and density diminish. Other risk factors, such as age and comorbidity that are also strongly correlated with skeletal muscle mass and density, become more important for the clinical course of a given patient. We hypothesize that most cancer populations, such as stage I-III colorectal

cancer patients, generally reach a functional status after treatment that is close to pre-disease levels [34]. In contrast, other cancer populations including sicker patients with an unfavorable prognosis and more dismal outcome, such as pancreatic or liver cancer patients, have an increased risk of spiraling down a vicious circle progressively enhancing physical impairment. Furthermore, tumor location, tumor biology, and tumor metabolism may determine the proportion of skeletal muscle wasting rather than tumor stage. Finally, our studies are limited by the fact that skeletal muscle mass was measured on one time point only, whereas skeletal muscle wasting is a process over time. Some studies showed that the loss of skeletal muscle mass during this period was more predictive of outcome than single measurements [15, 35, 36]. Moreover, anti-cancer surgery may also halt further skeletal muscle wasting.

A recent study found that advanced age, female gender, higher ASA classification, and an altered systemic inflammatory response were significantly associated with loss of skeletal muscle mass over time after colorectal surgery [37]. However, its prognostic value was not described and consequently remains unknown. Postoperative skeletal muscle depletion after resection for hepatocellular carcinoma did indeed identify patients with recurrence [38]. Future studies among patients with other tumors should therefore also elaborate on the question whether anticancer treatment may halt muscle wasting, and if postoperative skeletal muscle mass loss, which could be measured on sequential CT examinations, may predict mortality or could be used as a biomarker to identify disease recurrence in an early phase. We believe that the majority of patients with colorectal cancer undergoing surgery almost fully recover. Nonetheless, particularly elderly show a permanent loss in physical capacity after colorectal cancer surgery [34]. Early physical activity after surgery, however, seems important to maintain physical status [39].

In **part III** of this thesis we investigated the consequences of sarcopenia in patients listed for or undergoing liver transplantation. First, we performed a systematic review and meta-analysis, which is described in **chapter 10.** We included 19 studies with a total of 3,803 patients. After pooling the data, a non-significant association between sarcopenia and waiting list mortality (HR 1.75, 95% CI, 0.99-3.00, p=0.05) and an independent association between sarcopenia and post-transplant survival (HR 1.84, 95% CI 1.11-3.05, p=0.02) was found, independent of Model for End-stage Liver Disease (MELD) score. Although the MELD score is a very strong predictor for waiting list mortality, we successfully validated the association between sarcopenia and waiting list mortality in all consecutive patients with cirrhosis listed for transplantation between 2007 and 2014 in the Netherlands in **chapter 11**. In the 585 included patients, we found a significantly shorter waiting list survival in patients with sarcopenia, particularly in patients with a low MELD score. A final model developed using competing risk analysis, included MELD,

sarcopenia, hepatic encephalopathy before listing, and age. However, incorporating sarcopenia in the MELD score to enhance prioritizing liver transplant candidates and allocating donor organs has limited added value. The discrepancy between our results and the results of previous studies may be explained by the fact that we used competing risk analysis rather than Cox regression analysis, which indeed resulted in a significantly shorter waiting list survival in our patients with sarcopenia. Furthermore, it may be attributed to the fact that prioritization and allocation of donor organs and transplant candidates as well as waiting list mortality are all strongly related to the MELD score. This may have led to a self-fulfilling prophecy.

All patients included in chapters 10 and 11 with hepatocellular carcinoma (HCC) underwent liver transplantation within the Milan criteria [40]. These criteria state that patients are selected for liver transplantation when there is a single lesion smaller than five centimeters, or when there are up to three lesions smaller than three centimeters, without any vascular invasion. Patients meeting the Milan criteria have a 5-year survival rate of at least 70% and recurrence incidence of only 10% [41, 42]. However, in recent years it has been argued that the Milan criteria are too restrictive. Although most patients with HCC beyond the Milan criteria may experience disease recurrence after liver transplantation leading to decreased 5-year survival rates (53.6% versus 73.3%) [42], selected patients may still benefit and reach a 5-year survival that is comparable with recipients fulfilling the Milan criteria. The latest proposal to select eligible patients is known as the up-to-seven rule: the sum of the number of nodule(s) and the maximum diameter of the nodule(s) must not exceed the value of seven which resulted in a 5-year overall survival of 70% inpatients transplanted beyond the Milan criteria [42]. Future studies may investigate the addition of skeletal muscle mass as a biomarker to improve identification of patients with HCC who may or may not benefit from transplantation.

The socio-economic consequences of low skeletal muscle mass are described in **part IV** of this thesis. Following a study from the United States that described significantly higher hospital costs in patients with sarcopenia after cancer surgery [43], we found significantly higher hospital expenditure in patients with sarcopenia undergoing abdominal cancer surgery in a Western-European healthcare system (**chapter 12**). Independently of age, sex, BMI, ASA classification, and the extent of surgery, total hospital costs in patients with sarcopenia were almost €5,000 higher compared with patients without sarcopenia (p=0.004). Low skeletal muscle mass, as a measure of patient frailty, may thus be used as a parameter for case-mix corrections to compare treatment outcomes between centers.

In **chapter 13** we found similar results for patients with cirrhosis listed for liver transplantation: the median total hospital costs for patients with sarcopenia were €11,294 (IQR 3,570-46,469) compared with €6,878 (IQR 1,305-20,683) in patients without sarcopenia (p=0.008). In multivariable regression analysis, an incremental increase in skeletal muscle index (SMI) was significantly associated with a decrease in total costs (€458 per incremental SMI, 95%CI 14-902, p=0.043), independent of the total time on the waiting list. Results are in line with previous reports on hospital costs and an underestimation rather than an overestimation. Based on these two studies, we conclude that sarcopenia is independently associated with increased health-related costs for patients undergoing cancer surgery and patients on the waiting list for liver transplantation. These results justify the use of resources to find ways to optimize skeletal muscle mass, which may lead to a decrease in hospital expenditure, in addition to greater health benefit for the patient in an era of incremental healthcare costs and an increasingly aging population. Future prospective studies regarding skeletal muscle mass should consider including total costs as outcome measure.

FUTURE PERSPECTIVES

Future studies are needed to search for strategies to treat skeletal muscle wasting, which should be multimodal. A combination of physical exercise and nutritional support with or without therapeutic to treat low skeletal muscle mass and to halt or reverse skeletal muscle wasting should be opted for. In patients undergoing surgery, future research should reveal whether this should preferably be performed pre- and/or postoperatively. Modernizing waiting list targets may be needed to enable preoperative rehabilitation, and consequently longer waiting lists, may be beneficial [44]. After all. it may lead to decreased postoperative morbidity and increased survival, as well as increased quality of life [45, 46] and decreased fatigue complaints [47]. Recently, serum levels of myostatin, a cytokine involved in the regulation of muscle protein synthesis, was identified to be correlated with skeletal muscle mass and overall survival in patients with liver cirrhosis [48]. Although these results have not been validated yet, myostatin seems a highly interesting and potential biomarker for further research. Myostatin not only seems a promising biomarker for skeletal muscle loss and survival prediction, but also is a potential target to develop therapies to halt or reverse skeletal muscle wasting. The study by Zhou *et al.*, showing that myostatin antagonism successfully reduces skeletal muscle depletion in a mouse model, is a key publication within the field of experimental skeletal muscle wasting research [49]. Currently, some phase II studies are being conducted to halt or reverse skeletal muscle wasting as part of the cancer cachexia syndrome [50].

In **chapter 14** we propose a study aimed to assess the efficacy of a patient-tailored, supervised 12-week rehabilitation program consisting of physical therapy, solution focused psychotherapy, and dietary optimization in surgically treated hepatopancreatobiliary cancer patients. The primary outcome will be fatigue following the intervention program, which is considered a clinically relevant and invalidating complaint in this population. Quality of life, which is greatly hampered in these patients [51-54], will be assessed. In addition, skeletal muscle mass and strength, physical fitness, overall survival, and cost-effectiveness are the other secondary outcome measures. Measurements will be repeated at six and twelve months after surgery to explore long term effects. Furthermore, the association between skeletal muscle mass and the other outcome measures will be investigated.

REFERENCES

1. Prado CM, Birdsell LA, Baracos VE. The emerging role of computerized tomography in assessing cancer cachexia. Curr Opin Support Palliat Care 2009; 3: 269-75.
2. Prado CM, Lieffers JR, McCargar LJ, Reiman T, Sawyer MB, Martin L, et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. Lancet Oncol 2008; 9: 629-35.
3. Huillard O, Mir O, Peyromaure M, Tlemsani C, Giroux J, Boudou-Rouquette P, et al. Sarcopenia and body mass index predict sunitinib-induced early dose-limiting toxicities in renal cancer patients. Br J Cancer 2013; 108: 1034-41.
4. Antoun S, Baracos VE, Birdsell L, Escudier B, Sawyer MB. Low body mass index and sarcopenia associated with dose-limiting toxicity of sorafenib in patients with renal cell carcinoma. Annals of oncology : official journal of the European Society for Medical Oncology / ESMO 2010; 21: 1594-8.
5. Antoun S, Borget I, Lanoy E. Impact of sarcopenia on the prognosis and treatment toxicities in patients diagnosed with cancer. Curr Opin Support Palliat Care 2013; 7: 383-9.
6. Levolger S, van Vugt JL, de Bruin RW, IJzermans JN. Systematic review of sarcopenia in patients operated on for gastrointestinal and hepatopancreatobiliary malignancies. Br J Surg 2015; 102: 1448-58.
7. Lee JS, He K, Harbaugh CM, Schaubel DE, Sonnenday CJ, Wang SC, et al. Frailty, core muscle size, and mortality in patients undergoing open abdominal aortic aneurysm repair. J Vasc Surg 2011; 53: 912-7.
8. Carey EJ. Sarcopenia in solid organ transplantation. Nutr Clin Pract 2014; 29: 159-70.
9. van Vugt JL, Levolger S, de Bruin RW, van Rosmalen J, Metselaar HJ, IJzermans JN. Systematic review and meta-analysis of the impact of computed tomography assessed skeletal muscle mass on outcome in patients awaiting or undergoing liver transplantation. Am J Transplant 2016.
10. Cakir H, Heus C, van der Ploeg TJ, Houdijk AP. Visceral obesity determined by CT scan and outcomes after colorectal surgery; a systematic review and meta-analysis. Int J Colorectal Dis 2015; 30: 875-82.
11. Cecchini S, Cavazzini E, Marchesi F, Sarli L, Roncoroni L. Computed tomography volumetric fat parameters versus body mass index for predicting short-term outcomes of colon surgery. World J Surg 2011; 35: 415-23.
12. Jung M, Volonte F, Buchs NC, Gayet-Ageron A, Pugin F, Gervaz P, et al. Perirenal fat surface area as a risk factor for morbidity after elective colorectal surgery. Dis Colon Rectum 2014; 57: 201-9.

13. van Vledder MG, Levolger S, Ayez N, Verhoef C, Tran TC, IJzermans JN. Body composition and outcome in patients undergoing resection of colorectal liver metastases. Br J Surg 2012; 99: 550-7.
14. Reisinger KW, van Vugt JL, Tegels JJ, Snijders C, Hulsewe KW, Hoofwijk AG, et al. Functional compromise reflected by sarcopenia, frailty, and nutritional depletion predicts adverse postoperative outcome after colorectal cancer surgery. Annals of surgery 2015; 261: 345-52.
15. van Grinsven J, van Vugt JL, Gharbharan A, Bollen TL, Besselink MG, van Santvoort HC, et al. The Association of Computed Tomography-Assessed Body Composition with Mortality in Patients with Necrotizing Pancreatitis. Journal of gastrointestinal surgery : official journal of the Society for Surgery of the Alimentary Tract 2017.
16. Giusto M, Lattanzi B, Albanese C, Galtieri A, Farcomeni A, Giannelli V, et al. Sarcopenia in liver cirrhosis: the role of computed tomography scan for the assessment of muscle mass compared with dual-energy X-ray absorptiometry and anthropometry. Eur J Gastroenterol Hepatol 2015; 27: 328-34.
17. van der Werf A, Dekker IM, Meijerink MR, Wierdsma NJ, de van der Schueren MAE, Langius JAE. Skeletal muscle analyses: agreement between non-contrast and contrast CT scan measurements of skeletal muscle area and mean muscle attenuation. Clin Physiol Funct Imaging 2017.
18. Baracos VE. Psoas as a sentinel muscle for sarcopenia: a flawed premise. J Cachexia Sarcopenia Muscle 2017.
19. Rutten IJ, Ubachs J, Kruitwagen RFPM, Beets-Tan RGH, Olde Damink SWM, van Gorp T. Abstracts of the 9th International Conference on Cachexia, Sarcopenia, and Muscle Wasting, Berlin, Germany, 10–11 December 2016 (part 2) (1-45: Psoas muscle measurements are inferior to total skeletal muscle measurements in the assessment of sarcopenia in ovarian cancer). J Cachexia Sarcopenia Muscle 2017; 8: 161-83.
20. Takahashi N, Sugimoto M, Psutka SP, Chen B, Moynagh MR, Carter RE. Validation study of a new semi-automated software program for CT body composition analysis. Abdom Radiol (NY) 2017.
21. Popuri K, Cobzas D, Esfandiari N, Baracos V, Jagersand M. Body Composition Assessment in Axial CT Images Using FEM-Based Automatic Segmentation of Skeletal Muscle. IEEE Trans Med Imaging 2016; 35: 512-20.
22. Coelen RJ, Wiggers JK, Nio CY, Besselink MG, Busch OR, Gouma DJ, et al. Preoperative computed tomography assessment of skeletal muscle mass is valuable in predicting outcomes following hepatectomy for perihilar cholangiocarcinoma. HPB (Oxford) 2015; 17: 520-8.
23. Martin L, Birdsell L, Macdonald N, Reiman T, Clandinin MT, McCargar LJ, et al. Cancer cachexia in the age of obesity: skeletal muscle depletion is a powerful prognostic factor, independent of body mass index. J Clin Oncol 2013; 31: 1539-47.

24. Lieffers JR, Bathe OF, Fassbender K, Winget M, Baracos VE. Sarcopenia is associated with postoperative infection and delayed recovery from colorectal cancer resection surgery. Br J Cancer 2012; 107: 931-6.

25. Krell RW, Kaul DR, Martin AR, Englesbe MJ, Sonnenday CJ, Cai S, et al. Association between sarcopenia and the risk of serious infection among adults undergoing liver transplantation. Liver Transpl 2013; 19: 1396-402.

26. Montano-Loza AJ, Meza-Junco J, Baracos VE, Prado CM, Ma M, Meeberg G, et al. Severe muscle depletion predicts postoperative length of stay but is not associated with survival after liver transplantation. Liver Transpl 2014; 20: 640-8.

27. Moyes LH, Leitch EF, McKee RF, Anderson JH, Horgan PG, McMillan DC. Preoperative systemic inflammation predicts postoperative infectious complications in patients undergoing curative resection for colorectal cancer. Br J Cancer 2009; 100: 1236-9.

28. Reisinger KW, Derikx JP, van Vugt JL, Von Meyenfeldt MF, Hulsewe KW, Olde Damink SW, et al. Sarcopenia is associated with an increased inflammatory response to surgery in colorectal cancer. Clin Nutr 2016; 35: 924-7.

29. Richards CH, Roxburgh CS, MacMillan MT, Isswiasi S, Robertson EG, Guthrie GK, et al. The relationships between body composition and the systemic inflammatory response in patients with primary operable colorectal cancer. PLoS One 2012; 7: e41883.

30. Makary MA, Segev DL, Pronovost PJ, Syin D, Bandeen-Roche K, Patel P, et al. Frailty as a predictor of surgical outcomes in older patients. Journal of the American College of Surgeons 2010; 210: 901-8.

31. Cabilan CJ, Hines S. The short-term impact of colorectal cancer treatment on physical activity, functional status and quality of life: a systematic review. JBI Database System Rev Implement Rep 2017; 15: 517-66.

32. van Abbema D, van Vuuren A, van den Berkmortel F, van den Akker M, Deckx L, Buntinx F, et al. Functional status decline in older patients with breast and colorectal cancer after cancer treatment: A prospective cohort study. Journal of geriatric oncology 2017; 8: 176-84.

33. Otsuji H, Yokoyama Y, Ebata T, Igami T, Sugawara G, Mizuno T, et al. Preoperative sarcopenia negatively impacts postoperative outcomes following major hepatectomy with extrahepatic bile duct resection. World J Surg 2015; 39: 1494-500.

34. Hamaker ME, Prins MC, Schiphorst AH, van Tuyl SA, Pronk A, van den Bos F. Long-term changes in physical capacity after colorectal cancer treatment. Journal of geriatric oncology 2015; 6: 153-64.

35. Reisinger KW, Bosmans JWAM, Uittenbogaart M, Alsoumali A, Poeze M, Sosef MN, et al. Loss of Skeletal Muscle Mass During Neoadjuvant Chemoradiotherapy Predicts Postoperative Mortality in Esophageal Cancer Surgery. Ann Surg Oncol 2015; 22: 4445-52.

36. Blauwhoff-Buskermolen S, Versteeg KS, de van der Schueren MA, den Braver NR, Berkhof J, Langius JA, et al. Loss of Muscle Mass During Chemotherapy Is Predictive for Poor Survival of Patients With Metastatic Colorectal Cancer. J Clin Oncol 2016; 34: 1339-44.

37. Malietzis G, Currie AC, Johns N, Fearon KC, Darzi A, Kennedy RH, et al. Skeletal Muscle Changes After Elective Colorectal Cancer Resection: A Longitudinal Study. Ann Surg Oncol 2016; 23: 2539-47.

38. Kobayashi A, Kaido T, Hamaguchi Y, Okumura S, Taura K, Hatano E, et al. Impact of postoperative changes in sarcopenic factors on outcomes after hepatectomy for hepatocellular carcinoma. J Hepatobiliary Pancreat Sci 2016; 23: 57-64.

39. van Zutphen M, Winkels RM, van Duijnhoven FJ, van Harten-Gerritsen SA, Kok DE, van Duijvendijk P, et al. An increase in physical activity after colorectal cancer surgery is associated with improved recovery of physical functioning: a prospective cohort study. BMC cancer 2017; 17: 74.

40. Mazzaferro V, Regalia E, Doci R, Andreola S, Pulvirenti A, Bozzetti F, et al. Liver transplantation for the treatment of small hepatocellular carcinomas in patients with cirrhosis. The New England journal of medicine 1996; 334: 693-9.

41. Mazzaferro V, Bhoori S, Sposito C, Bongini M, Langer M, Miceli R, et al. Milan criteria in liver transplantation for hepatocellular carcinoma: an evidence-based analysis of 15 years of experience. Liver Transpl 2011; 17 Suppl 2: S44-57.

42. Mazzaferro V, Llovet JM, Miceli R, Bhoori S, Schiavo M, Mariani L, et al. Predicting survival after liver transplantation in patients with hepatocellular carcinoma beyond the Milan criteria: a retrospective, exploratory analysis. Lancet Oncol 2009; 10: 35-43.

43. Gani F, Buettner S, Margonis GA, Sasaki K, Wagner D, Kim Y, et al. Sarcopenia predicts costs among patients undergoing major abdominal operations. Surgery 2016.

44. Sothisrihari SR, Wright C, Hammond T. Should preoperative optimization of colorectal cancer patients supersede the demands of the 62-day pathway? Colorectal Dis 2017; 19: 617-20.

45. Blauwhoff-Buskermolen S, Langius JAE, Becker A, Verheul HMW, de van der Schueren MAE. The influence of different muscle mass measurements on the diagnosis of cancer cachexia. J Cachexia Sarcopenia Muscle 2017.

46. Bye A, Sjoblom B, Wentzel-Larsen T, Gronberg BH, Baracos VE, Hjermstad MJ, et al. Muscle mass and association to quality of life in non-small cell lung cancer patients. J Cachexia Sarcopenia Muscle 2017.

47. Neefjes ECW, van den Hurk RM, Blauwhoff-Buskermolen S, van der Vorst M, Becker-Commissaris A, de van der Schueren MAE, et al. Muscle mass as a target to reduce fatigue in patients with advanced cancer. J Cachexia Sarcopenia Muscle 2017.

48. Nishikawa H, Enomoto H, Ishii A, Iwata Y, Miyamoto Y, Ishii N, et al. Elevated serum myostatin level is associated with worse survival in patients with liver cirrhosis. J Cachexia Sarcopenia Muscle 2017.

49. Zhou X, Wang JL, Lu J, Song Y, Kwak KS, Jiao Q, et al. Reversal of cancer cachexia and muscle wasting by ActRIIB antagonism leads to prolonged survival. Cell 2010; 142: 531-43.

50. Dingemans AM, de Vos-Geelen J, Langen R, Schols AM. Phase II drugs that are currently in development for the treatment of cachexia. Expert Opin Investig Drugs 2014; 23: 1655-69.

51. Uitdehaag MJ, Verschuur EM, van Eijck CH, van der Gaast A, van der Rijt CC, de Man RA, et al. Problems and needs in patients with incurable esophageal and pancreaticobiliary cancer: a descriptive study. Gastroenterol Nurs 2015; 38: 42-54.

52. Morak MJ, Pek CJ, Kompanje EJ, Hop WC, Kazemier G, van Eijck CH. Quality of life after adjuvant intra-arterial chemotherapy and radiotherapy versus surgery alone in resectable pancreatic and periampullary cancer: a prospective randomized controlled study. Cancer 2010; 116: 830-6.

53. Huang G, Chen X, Lau WY, Shen F, Wang RY, Yuan SX, et al. Quality of life after surgical resection compared with radiofrequency ablation for small hepatocellular carcinomas. Br J Surg 2014; 101: 1006-15.

54. Meier A, Yopp A, Mok H, Kandunoori P, Tiro J, Singal AG. Role functioning is associated with survival in patients with hepatocellular carcinoma. Qual Life Res 2015; 24: 1669-75.

NEDERLANDSE SAMENVATTING

Met name in een periode van toenemende vergrijzing, waarin kanker een van de belangrijkste doodsoorzaken is en het tekort aan donororganen toeneemt, zou skeletspiermassa gemeten op computed tomography (CT) van toegevoegde waarde kunnen zijn om de fysiologische reserves van een patiënt op een objectieve manier in te schatten. Daarom onderzochten wij de lange- en korte termijn effecten van lage skeletspiermassa in chirurgische patiënten, welke beschreven staan in deze dissertatie.

In **deel I** van dit proefschrift onderzochten we de accuratesse van verschillende methoden voor het meten van skeletspiermassa en - dichtheid op CT afbeeldingen. De afgelopen jaren zijn er vele studies uitgevoerd waarbij spiermassa gemeten is op CT in verschillende populaties, zoals medisch- of chirurgisch oncologische, vaatchirurgische en transplantatie patiënten. Daarnaast werd tevens vaak vetmassa gemeten op CT afbeeldingen. Hiervoor werden echter verschillende soorten software gebruikt, zoals FatSeg, OsiriX, ImageJ en SliceOmatic. Om studieresultaten te kunnen vergelijken, hebben we deze verschillende software in **hoofdstuk 2** met elkaar vergeleken. Uit de resultaten bleek dat de metingen verricht met verschillende soorten software en uitgevoerd door verschillende onderzoekers uitstekend overeen kwamen.

Hoewel CT als de gouden standaard beschouwd wordt voor het meten van lichaamssamenstelling (dat wil zeggen: spiermassa en vetmassa), bestaat er momenteel geen gestandaardiseerde methode om deze metingen te verrichten. De invloed van intraveneus contrast, bijvoorbeeld, was onbekend. Daarom hebben we in **hoofdstuk 3** de invloed van intraveneus contrast op de gemeten hoeveelheid spiermassa en de dichtheid van de spiermassa onderzocht. De resultaten toonden dat intraveneus contrast geen invloed heeft op de gemeten hoeveelheid spiermassa, maar wel op de gemeten dichtheid. Welke contrast-fase gebruikt wordt, maakt echter niet uit. Daarom pleiten wij voor het gebruik van de (porto)veneuze contrastfase, aangezien deze bij kanker- en levertransplantatie patiënten het meest voorhanden is.

Omdat er tot op heden veel discussie bestaat over gebruikte afkapwaarden om patiënten in te delen in groepen met hoge- dan wel lage spiermassa en er weinig bekend is over op CT gemeten spiermassa in gezonde personen, hebben we in **hoofdstuk 4** spiermassa en spierdichtheid gemeten in ruim 1.000 levende nier donoren. Levende nierdonoren dienen niet als patiënt beschouwd te worden, maar als 'gezonde vrijwilliger'. De hoeveelheid spiermassa en de spierdichtheid waren sterk afhankelijk van geslacht,

leeftijd en body mass index (BMI). Op basis van de gevonden resultaten hebben we een nomogram ontwikkeld, waarmee de geschatte gezonde spiermassa- en dichtheid van een patiënt berekend kunnen worden.

De impact van lage skeletspiermassa en –dichtheid voor chirurgisch-oncologische patiënten wordt beschreven in **deel II** van dit proefschrift. In **hoofdstuk 5** hebben we een systematische review verricht waarbij we de impact van lage skeletspiermassa op postoperatieve uitkomsten in gastro-intestinale en hepatopancreatobiliaire chirurgie beschrijven op basis van de huidige literatuur. Er werden in totaal 13 studies met 2.884 patiënten geïncludeerd. Met name in patiënten die chirurgie ondergingen van colorectale kanker werd een associatie gevonden tussen lage spiermassa en korte termijn postoperatieve uitkomsten. We vonden een grote variatie in gebruikte methoden voor het meten van skeletspiermassa en het definiëren van lage skeletspiermassa, waardoor we niet in staat waren een meta-analyse van de resultaten te verrichten.

In **hoofdstuk 6** beschrijven we de associatie tussen verminderde weerbaarheid (gedefinieerd als lage skeletspiermassa, kwetsbaarheid ('frailty') en ondervoeding) en uitkomsten na chirurgie vanwege colorectaal carcinoom. Mortaliteit was significant hoger in patiënten met lage skeletspiermassa vergeleken met patiënten met een hoge skeletspiermassa. Hoewel lage skeletspiermassa niet geassocieerd was met postoperatieve naadlekkage en sepsis, was de combinatie van lage skeletspiermassa, kwetsbaarheid en ondervoeding sterk voorspellend voor het optreden van postoperatieve sepsis.

In de twee daarop volgende hoofdstukken hebben we de resultaten in colorectaal carcinoom patiënten gevalideerd. Allereerst vonden we in **hoofdstuk 7** een significante associatie tussen de hoeveelheid skeletspiermassa en het optreden van ernstige postoperatieve complicaties na cytoreductieve chirurgie in combinatie met hyperthermische intraperitioneale chemotherapie (HIPEC). Vervolgens vonden we in **hoofdstuk 8** in een groot, multicentrisch cohort van stadium I-III colorectaal carcinoom patiënten een significante associatie tussen lage skeletspiermassa en postoperatieve complicaties. Een associatie met lange termijn ziektevrije-, kanker specifieke- of algemene overleving vonden we niet. In **hoofdstuk 9** vonden we een significant kortere algemene overleving in patiënten met perihilair cholangiocarcinoom met een lage skeletspierdichtheid vergeleken met patiënten met een hoge skeletspierdichtheid, ongeacht het soort therapie dat de patiënten onderging. Een effect van skeletspiermassa op overleving werd niet gevonden.

In **deel III** onderzochten we het effect van lage skeletspiermassa in levertransplantatie patiënten. In **hoofdstuk 10** beschrijven we de resultaten van een systematisch review en meta-analyse waarin 19 studies met een totaal van 3.803 patiënten werden geïncludeerd. De meta-analyse van de gepoolde data resulteerde in een niet-significante associatie tussen lage skeletspiermassa en mortaliteit op de wachtlijst voor levertransplantatie en een significante associatie tussen lage skeletspiermassa en overleving na levertransplantatie. Dit laatste effect was onafhankelijk van de MELD-score. Zoals bij de chirurgisch-oncologische studies, vonden we ook hier een grote diversiteit aan methodiek. Bovendien was een groot deel van de studies verricht in overlappende studiepopulaties. Daarom onderzochten we in een cohort van alle patiënten die in Nederland tussen 2007 en 2014 op de wachtlijst geplaatst werden voor levertransplantatie vanwege levercirrose, zoals beschreven in **hoofdstuk 11**, de associatie tussen lage skeletspiermassa en wachtlijst mortaliteit middels een competing risk analyse. We vonden een significant lagere wachtlijst overleving in patiënten met een lage skeletspiermassa. Lage skeletspiermassa had echter geen toegevoegde waarde op de MELD-score voor het voorspellen van wachtlijst mortaliteit.

De socio-economische consequenties van lage skeletspiermassa worden beschreven in **deel IV** van deze dissertatie. In **hoofdstuk 12** beschrijven we een studie naar de associatie tussen preoperatieve spiermassa en perioperatieve ziekenhuiskosten in patiënten die chirurgie ondergaan vanwege gastro-intestinale maligniteiten. Uit de resultaten bleek dat de ziekenhuiskosten bijna €5.000 hoger waren in patiënten met een lage skeletspiermassa vergeleken met patiënten met een hoge skeletspiermassa, waarbij gecorrigeerd werd voor leeftijd, geslacht, BMI, ASA classificatie en de grootte van de ingreep. Vergelijkbare resultaten werden gevonden in de studie beschreven in **hoofdstuk 13**, waarin we hebben onderzocht wat het effect is van lage skeletspiermassa op de ziekenhuiskosten ten tijde van wachtlijst plaatsing voor levertransplantatie. Een toenemende skeletspiermassa was onafhankelijk geassocieerd met een afname in totale ziekenhuiskosten (€458 per toegenomen cm^2/m^2 van de skeletal muscle index). Dit effect was onafhankelijk van bijvoorbeeld de totale periode dat een patiënt op de wachtlijst geplaatst stond.

In **hoofdstuk 14** wordt het protocol van een gerandomiseerde studie beschreven waarin we het effect van een rehabilitatieprogramma - bestaande uit fysiotherapie, psychosociale therapie en voedingsadviezen – zullen onderzoeken op vermoeidheidsklachten bij patiënten die chirurgie ondergaan vanwege hepatopancreatobiliaire maligniteiten. Ernstige vermoeidheid komt veel voor bij deze groep patiënten en heeft invaliderende gevolgen die leiden tot een verminderde kwaliteit van leven. Daarom zal ook het effect op kwaliteit van leven onderzocht worden, evenals het effect op spiermassa, spierkracht

en de fysieke conditie. Ook het effect op overleving en de kosteneffectiviteit van het programma zullen bepaald worden. Indien dit programma succesvol blijkt, zullen wij streven naar implementatie hiervan in de reguliere zorg.

APPENDICES

A

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A

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LIST OF PUBLICATIONS

In this thesis

Estimated Skeletal Muscle Mass and Density Values Measured on Computed Tomography Examinations in over One Thousand Healthy Subjects.
van Vugt JL, van Putten Y, van der Kall IM, Buettner S, de Bruin RW, Dekker HM, d'Ancona FC, Kimenai HJ, Warlé MC, IJzermans JN.
Submitted.

Impact of Low Skeletal Muscle Mass and Density on Short and Long-Term Outcome After Resection of Stage I-III Colorectal Cancer: Results From a Prospective Multicenter Observational Study.
van Vugt JL, Coebergh van den Braak RR, Lalmahomed ZS, Vrijland WW, Dekker JW, Zimmerman DD, Vles WJ, Coene PP, IJzermans JN.
Submitted.

Low Skeletal Muscle Density is Associated with Early Death in Patients with Perihilar Cholangiocarcinoma.
van Vugt JL, Gaspersz MP, Vugts J, Buettner S, Levolger S, de Bruin RW, Polak WG, de Jonge J, Willemssen FE, Groot Koerkamp B, IJzermans JN.
Submitted.

A Nomogram Including Sarcopenia Surpasses the MELD Score in Predicting Waiting List Mortality in Cirrhotic Liver Transplant Candidates: A Competing Risk Analysis in a National Cohort.
van Vugt JL, Alferink LJ, Buettner S, Gaspersz M, Bot D, Darwish Murad S, Feshtali S, van Ooijen PM, Polak WG, Porte RJ, van Hoek B, van den Berg AP, Metselaar HJ, IJzermans JN.
Journal of Hepatology.

Sarcopenia is Associated with Increased Hospital Expenditure in Patients Undergoing Cancer Surgery of the Alimentary Tract.
van Vugt JL, Buettner S, Levolger S, Coebergh van den Braak RR, Suker M, Gaspersz MP, de Bruin RW, Verhoef C, van Eijck CH, Bossche N, Groot Koerkamp B, IJzermans JN.
PloS One 2017 Oct 31; 12 (10): e0186547.

Low Skeletal Muscle Mass is Associated With Increased Hospital Costs in Patients with Cirrhosis Listed for Liver Transplantation.
van Vugt JL, Buettner S, Alferink LJ, Bossche N, de Bruin RW, Darwish Murad S, Polak WG, Metselaar HJ, IJzermans JN.
Transplant International 2017 Sep 4 [Epub ahead of print].

Contrast-enhancement Influences Skeletal Muscle Density, but not Skeletal Muscle Mass, Measurements on Computed Tomography.
van Vugt JL, Coebergh van den Braak RR, Schippers HJ, van Veen K, Levolger S, de Bruin RW, Koek M, Niessen WJ, IJzermans JN, Willemssen FE.
Clinical Nutrition 2017 Jul 14 [Epub ahead of print].

A Comparative Study of Software Programs for Cross-Sectional Skeletal Muscle and Adipose Tissue Measurements on Abdominal Computed Tomography Scans of Rectal Cancer Patients.
van Vugt JL, Levolger S, Gharbharan A, Koek M, Niessen WJ, Burger JW, Willemsen SP, de Bruin RW, IJzermans JN.
Journal of Cachexia, Sarcopenia and Muscle 2017 Apr; 8(2):285-297.

Systematic Review and Meta-Analysis of the Impact of Computed Tomography Assessed Skeletal Muscle Mass in Patients Awaiting or Undergoing Liver Transplantation.
van Vugt JL*, Levolger S*, de Bruin RW, van Rosmalen J, Metselaar HJ, IJzermans JN.
American Journal of Transplantation 2016 Aug; 16(8):2277-92.

Skeletal Muscle Depletion is Associated with Severe Postoperative Complications in Patients Undergoing Cytoreductive Surgery with HIPEC for Peritoneal Carcinomatosis of Colorectal Cancer.
van Vugt JL, Braam HJ, van Oudheusden TR, Vestering A, Bollen TL, Wiezer MJ, de Hingh IH, van Ramshorst B, Boerma D.
Annals of Surgical Oncology 2015 Oct; 22(11):3625-31.

Systematic Review of Sarcopenia in Patients Operated on for Gastrointestinal and Hepatopancreatobiliary Malignancies.
Levolger S*, **van Vugt JL***, de Bruin RW, IJzermans JN.
British Journal of Surgery 2015 Nov; 102(12):1448-58.

Functional Compromise Reflected by Sarcopenia, Frailty and Nutritional Depletion Predicts Adverse Postoperative Outcome after Colorectal Cancer Surgery.
Reisinger KW, **van Vugt JL**, Tegels JJ, Snijders C, Hulsewé KW, Hoofwijk AG, Stoot JH, von Meyenfeldt, MF, Beets GL, Derikx, JP, Poeze M.
Annals of Surgery 2015 Feb; 261(2):345-352.

Not in this thesis

Skeletal Muscle Mass, Delirium Risk Factor in Elderly Patients after Colorectal Cancer Surgery.
Mosk CA, **van Vugt JL**, de Jonge H, Witjes C, Buettner S, IJzermans JN, van der Laan L.
Submitted.

Translating the ABC-02 Trial into Daily Practice: Outcome of Palliative Treatment in Patients with Unresectable Biliary Tract Cancer Treated with Gemcitabine and Cisplatin.
Dierks J, Gaspersz MP, Belkouz A, **van Vugt JL**, Coelen RJ, de Groot JW, ten Tije AJ, Meijer WG, Pruijt JF, van Voorthuizen T, van Spronsen DJ, Rentinck M, ten Oever D, Smit JM, Otten HM, van Gulik TM, Wilmink JW, Groot Koerkamp B, Klümpen HJ.
Submitted.

The New Body Mass Index Formula; Not Validated as a Predictor of Outcome in a Large Cohort Study of Patients Undergoing General Surgery.
Tjeertes EK, Hoeks SE, **van Vugt JL**, Stolker RJ, Hoofwijk AG.
Clinical Nutrition 2017 (In press).

The Prognostic Value of Hepatic Artery and Portal Vein Involvement in Patients with Perihilar Cholangiocarcinoma.
van Vugt JL*, Gaspersz MP*, Coelen RJ, Vugts J, Labeur TA, de Jonge J, Polak WG, IJzermans JN, Nio CY, van Gulik TM, Willemssen FE*, Groot Koerkamp B*.
HPB (Oxford) 2017 Sep 25 [Epub ahead of print].

The First Year after Colorectal Surgery in the Elderly.
Kornmann VN, **van Vugt JL**, Smits AB, van Ramshorst B, Boerma D.
Annals of Coloproctology 2017 Aug; 33(4):134-138.

Conditional Probability of Long-term Survival in Patients with Locally Advanced and Metastatic Hilar Cholangiocacinoma.
Gaspersz MP, Buettner S, **van Vugt JL**, Roos E, Coelen RJ, van Gulik TM, IJzermans JN, Groot Koerkamp B.
HPB (Oxford) 2017 Nov; 19(11):966-971.

Performance of Prognostic Scores and Staging Systems in Predicting Long-Term Survival Outcomes after Surgery for Intrahepatic Cholangiocarcinoma.
Buettner S, Galjart B, **van Vugt JL**, Bagante F, Alexandrescu S, Marques HP, Lamelas J, Aldrighetti L, Gamblin TC, Maithel SK, Pulitano C, Margonis GA, Weiss M, Bauer TW, Shen F, Poultsides GA, Marsh JW, IJzermans JN, Groot Koerkamp B, Pawlik TM.
Journal of Surgical Oncology 2017 Jul 13 [Epub ahead of print].

Anatomical Resections Improve Disease Free Survival in Patients with *KRAS*-Mutated Colorectal Liver Metastases.
Margonis GA, Buettner S, Andreatos N, Sasaki K, IJzermans JN, **van Vugt JL**, Cameron JL, He J, Wolfgang CL, Weiss MJ.
Annals of Surgery 2017 Oct; 266(4):641-649.

Survival after Resection of Perihilar Cholangiocarcinoma in Patients with Lymph Node Metastases.
Buettner S, **van Vugt JL**, Gaspersz MP, Coelen RJ, Labeur TA, Ethun CG, Maithel SK, Poultsides G, Tran T, Idrees K, Isom CA, Fields RC, Krasnick B, Weber SM, Salem A, Martin RC, Scoggins C, Shen P, Mogal HD, Schmidt C, Beal E, Hatzaras I, Shenoy R, IJzermans JN, van Gulik TM, Groot Koerkamp B, Pawlik TM.
HPB (Oxford) 2017 Aug; 19(8):735-740.

Validation of the Mayo Clinic Staging System in Determining Prognoses of Patients with Perihilar Cholangiocarcinoma.
Coelen RJ, Gaspersz MP, Labeur TA, **van Vugt JL**, van Dieren S, Willemssen FE, Nio CY, IJzermans JN, Klümpen HJ, Groot Koerkamp B, van Gulik TM.
Clinical Gastroenterology and Hepatology 2017 May 19 [Epub ahead of print].

The Association of Computed Tomography Assessed Body Composition Analysis and Mortality in Patients with Necrotizing Pancreatitis.
van Grinsven J*, **van Vugt JL***, Gharbharan A, Bollen TL, Besselink MG, van Santvoort HC, van Eijck CH, Boerma D.
Journal of Gastrointestinal Surgery 2017 Jun; 21(6):1000-1008.

Intrahepatic Cholangiocarcinoma: Current Perspectives.
Buettner S, **van Vugt JL**, IJzermans JN, Groot Koerkamp B.
OncoTargets and Therapy 2017 Feb; 10:1131-1142.

A Comparison of Prognostic Schemes for Perihilar Cholangiocarcinoma.
Buettner S, **van Vugt JL**, Gani F, Groot Koerkamp B, Margonis GA, Ethun CG, Poultsides G, Tran T, Idrees K, Isom CA, Fields RC, Krasnick B, Weber SM, Salem A, Martin RC, Scoggins C, Shen P, Mogal HD, Schmidt C, Beal E, Hatzaras I, Shenoy R, Maithel SK, Guglielmi A, IJzermans JN, Pawlik TM.
Journal of Gastrointestinal Surgery 2016 Oct; 20(10):1716-24.

Reply to: Comparing the Variability between Measurements for Sarcopenia Using Magnetic Resonance Imaging and Computed Tomography Imaging.
van Vugt JL, Levolger S, Metselaar HJ, IJzermans JN.
American Journal of Transplantation 2016 Sep; 16(9):2768 (Letter to the Editor).

Sarcopenia is Associated With an Increased Inflammatory Response to Surgery in Colorectal Cancer.
Reisinger KW, Derikx JP, **van Vugt JL**, Von Meyenfeldt MF, Hulsewé KW, Olde Damink SW, Stoot JH, Poeze M.
Clinical Nutritrion 2016 Aug; 35(4):924-7.

Sidestream Dark Field Imaging of the Serosal Microcirculation During Gastrointestinal Surgery.
de Bruin AF, Kornmann VN, van der Sloot K, **van Vugt JL**, Gosselink MP, Smits AB, van Ramshorst B, Boerma EC, Noordzij PG, Boerma D, van Iterson M.
Colorectal Disease 2016 Mar; 18(3):O103-10.

Reply on: The New Body Mass Index as a Predictor of Postoperative Complications in Elective Colorectal Cancer Surgery – Letter to the Editor.
van Vugt JL, Cakir H, Kornmann VN, Doodeman HJ, Stoot JH, Boerma D, Houdijk AP, Hulsewé KW.
Clinical Nutrition 2016 Feb; 11:e71 (Letter to the Editor).

The Increasing Use of Computed Tomography to Assess Body Composition and its Clinical Relevance.
Trenning TA, Gharbharan A, Levolger S, **van Vugt JL**.
Erasmus Journal of Medicine 2015 Oct; 5(1):51-54.

Sarcopenia is Highly Prevalent in Patients Undergoing Surgery for Gastric Cancer but not Associated with Worse Outcomes.
Tegels JJ, **van Vugt JL**, Reisinger KW, Hulsewé KW, Hoofwijk AG, Derikx JP, Stoot JH.
Journal of Surgical Oncology 2015 Sep; 112(4);403-7.

The Impact of Sarcopenia on Survival and Complications in Surgical Oncology: A Review of the Current Literature.
van Vugt JL, Levolger S, Coelen RJ, de Bruin RW, IJzermans JN.
Journal of Surgical Oncology 2015 Nov; 112(6):681-2 (Letter to the Editor).
Elevated High-Sensitive Cardiac Troponin T Levels are Associated with Low Skeletal

Muscle Mass in Abdominal Surgical Oncology Patients at Risk for Coronary Artery Disease.
van Vugt JL, Boerma D, Dijkstra IM, Bollen TL, Eefting FD, IJzermans JN, Noordzij PG.
International Journal of Cardiology 2015 Jul 15; 191:229-30.

Nasal Carriage of *Staphylococcus aureus* Among Surgeons and Surgical Residents: A Nationwide Prevalence Study.
van Vugt JL*, Coelen RJ*, van Dam DW, Winkens B, Derikx JP, Heddema ER, Stoot JH.
Surgical Infections (Larchmt) 2015 Apr; 16(2):178-82.

First Experiences with the Radial Reload with Tristaple Technology™ in Low Rectal Surgery.
van Vugt JL, Tegels JJ, Derikx JP, Visschers RG, Stoot JH.
International Journal of Surgery 2015 Feb; 14:23-7.

Een enkel-voet letsel op vier niveaus.
van Vugt JL, van Rappard JR, Hammacher ER, Segers MJ.
Nederlands Tijdschrift voor Traumachirurgie 2014; 22(5);122-124.

Improving the Outcomes in Oncological Colorectal Surgery.
van Vugt JL, Reisinger KW, Derikx JP, Boerma D, Stoot JH.
World Journal of Gastroenterology 2014 Sep 21; 20(35):12445-12457.

The New Body Mass Index as a Predictor of Postoperative Complications in Elective Colorectal Cancer Surgery.
van Vugt JL, Cakir H, Kornmann VN, Doodeman HJ, Stoot JH, Boerma D, Houdijk AP, Hulsewé KW.
Clinical Nutrition 2015 Aug; 34(4):700-4.

De Morel-Lavallée-Laesie: Late klachten na een trauma.
van Vugt JL, Beks BJ, Borghans RA, Hoofwijk AG.
Nederlands Tijdschrift voor Geneeskunde 2013; 157:A5914.

Improving Outcome in Gastrointestinal and Hepatopancreaticobiliary Surgical Oncology by Preoperative Risk Assessment and Optimization of Perioperative Care.
Jongen AC*, van Woerden V*, **van Vugt JL**, de Hoogt PA, de Wijkerslooth de Weerdesteijn EM, Tegels JJ, Bouvy ND, Stoot JH.
Book chapter in: Oncology Critical Care, Jeffrey Hoag (Ed.), ISBN: 978-953-51-2782-6, InTech, DOI: 10.5772/64775, Available from: http://www.intechopen.com/books/oncology-critical-care/improving-outcomes-in-gastrointestinal-and-hepatopancreaticobiliary-surgical-oncology-by-preoperative.

General Introduction: Advances in Hepatic Surgery.
Stoot JH, Coelen RJ, **van Vugt JL**, Dejong CH.
Book chapter in: Hepatic Surgery, Prof. Hesham Abdeldayem (Ed.), InTech, ISBN: 978-953-51-0965-5, InTech, DOI: 10.5772/54710. Available from: http://www.intechopen.com/books/hepatic-surgery/general-introduction-advances-in-hepatic-surgery.

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ABOUT THE AUTHOR

The author of this thesis, Jeroen Laurens Ad van Vugt, was born on June 6, 1988 in Mierlo, the Netherlands. He graduated from the Carolus Borromeus College (VWO Gymnasium) in Helmond, the Netherlands, and started Medical School at Maastricht University (Maastricht, the Netherlands) in 2006. Besides being part of an exchange program with the University of Ferrara (Ferrara, Italy), he undertook internships in Belgium, India, and South-Africa. The first steps for his PhD thesis were taken during his clinical and scientific pre-graduating rotations at the Department of Surgery of the Orbis Medical Center (Sittard, the Netherlands; currently Zuyderland Medical Center), where he started research on the effect of skeletal muscle depletion in surgical cancer patients (supervisors dr. J.H.M.B. Stoot, dr. K.W.E. Hulsewé). This resulted in multiple scientific publications. After obtaining his medical degree in January 2013, he started as a resident not in training (ANIOS) and research fellow at the Department of Surgery of the St Antonius Hospital, Nieuwegein, the Netherlands (supervisors dr. P.M.N.Y.H. Go, dr. D. Boerma). He continued his research activities at the Erasmus MC University Medical Center, Rotterdam, the Netherlands (supervisors prof. J.N.M. IJzermans, dr. R.W.F. de Bruin, and dr. B. Groot Koerkamp), which eventually resulted in this work. Currently, he works as a resident at the Department of Surgery of the IJsselland Hospital (Capelle aan den IJssel, the Netherlands) under the supervision of dr. I. Dawson, where he will start his surgical traineeship in January 2018.

PHD PORTFOLIO SUMMARY

Summary of PhD training and teaching activities

Name PhD student:	J.L.A. van Vugt	Promotor:	Prof.dr. J.N.M. IJzermans
Erasmus MC Department:	Surgery	Supervisors:	dr. R.W.F. de Bruin,
PhD period:	2013 - 2017		dr. B. Groot Koerkamp
		Date of thesis defence:	20 December 2017

PhD training

Courses		Year	Workload (ECTS)
-	ICH-GCP (Good Clinical Practice)	2014	0.3
-	Biomedical English writing	2014	2.0
-	Research integrity	2015	0.3
-	Laboratory animal science	2015	0.3
-	Basic introduction in SPSS	2015	0.3
-	Introduction in GraphPad Prism	2015	0.3
-	Microsoft Excel	2015	0.3
-	Survival analysis	2015	0.5
Conferences			
-	Symposium Experimenteel Onderzoek Heelkundige Specialismen (SEOHS), Maastricht, the Netherlands	2013	1.0
-	International Surgical Week, Helsinki, Finland	2013	1.0
-	Nederlandse Vereniging voor Heelkunde (NVvH) Chirurgendagen, Veldhoven, the Netherlands	2013-2016	4.0
-	Nederlandse Vereniging voor Gastro-Enterologie (NVGE) Najaarscongres, Veldhoven, the Netherlands	2014-2015	2.0
-	8th International Conference on Cachexia, Sarcopenia and Muscle Wasting, Paris, France	2015	1.0
-	The Liver Meeting (AASLD), Boston, USA	2016	1.0
-	9th International Conference on Cachexia, Sarcopenia and Muscle Wasting, Berlin, Germany	2016	1.0
-	Americas Hepato-Pancreato-Biliary Association (AHPBA) Annual Meeting, Miami, USA	2017	1.0

Conferences	Year	Workload (ECTS)
- The International Liver Congress (EASL), Amsterdam, the Netherlands	2017	1.0
- 52nd Congress of the European Society for Surgical Research (ESSR), Amsterdam, the Netherlands	2017	1.0
- 18th Congress of the European Society for Organ Transplantation (ESOT), Barcelona, Spain	2017	1.0
- 10th International Conference on Cachexia, Sarcopenia and Muscle Wasting, Rome, Italy	2017	1.0
Podium presentations		
- International Surgical Week, Helsinki, Finland	2013	1.0
- Nederlandse Vereniging voor Heelkunde (NVvH) Chirurgendagen, Veldhoven, the Netherlands	2013-2014	2.0
- St Antonius Hospital Research Meeting, Nieuwegein, the Netherlands	2014	1.0
- Nederlandse Vereniging voor Gastro-Enterologie (NVGE) Najaarscongres, Veldhoven, the Netherlands	2014-2015	2.0
- Wetenschapsdag Heelkunde, Rotterdam, the Netherlands	2015	1.0
- Americas Hepato-Pancreato-Biliary Association (AHPBA) Annual Meeting, Miami, USA	2017	2.0
- 52nd Congress of the European Society for Surgical Research (ESSR), Amsterdam, the Netherlands	2017	1.0
- 18th Congress of the European Society for Organ Transplantation (ESOT), Barcelona, Spain	2017	1.0
- 10th International Conference on Cachexia, Sarcopenia and Muscle Wasting, Rome, Italy	2017	1.0
Seminars and workshops		
- Medical Business Masterclass (MBM), Amsterdam, the Netherlands	2013	1.0
- LETIS Lustrum Day, Rotterdam, the Netherlands	2015	0.3
- 19th Molecular Medicine Day, Rotterdam, the Netherlands	2015	0.3
- 2e Studiedag Voeding, Bewegen en Kanker, Utrecht, the Netherlands	2015	0.3
- 31st Erasmus Liver Day, Rotterdam, the Netherlands	2016	0.3
- Wetenschapsdag Heelkunde, Rotterdam, the Netherlands	2015-2017	1.0
- Value in Care Masterclass, Amsterdam, the Netherlands	2017	2.0

Teaching activities

		Year	Workload (ECTS)
Lecturing			
-	Tutoring first year medical students	2015	1.5
Supervising			
-	Basic life support examinations first year medical students	2015-2016	0.6
-	Master's thesis sarcopenia and colorectal cancer (2 students)	2015-2016	4.0
-	Master's thesis sarcopenia and necrotizing pancreatitis	2015	2.0
Other			
-	Supervising Bachelor's thesis	2016	2.0
-	Supervising Bachelor students	2016-2017	1.5
-	Reviewer for KWF Kankerbestrijding and scientific journals (British Journal of Nutrition, Clinical Nutrition, Colorectal Disease, Journal of Geriatric Oncology, Journal of Surgical Oncology, Nutrition, Plos One, Sarcoma, Transplantation, Transplant International)	2015-2017	4.0
Total ECTS:			**53.1**