Technische Universität München
TUM School of Medicine and Health

Skeletal Muscle Ultrasound and Functional Independence in Critically Ill Patients

Hugo Lanz

Vollständiger Abdruck der von der TUM School of Medicine and Health der Technischen Universität München zur Erlangung eines

Doktors der Medizin (Dr. med.)

genehmigten Dissertation.

Vorsitz: Prof. Dr. Marcus Makowski

Prüfende der Dissertation:

1. apl. Prof. Dr. Manfred Blobner
2. Priv.-Doz. Dr. Marc Hanschen

Die Dissertation wurde am 18.04.2023 bei der Technischen Universität München eingereicht und durch die TUM School of Medicine and Health am 08.11.2023 angenommen.

Acknowledgements

I would like to thank Prof. Dr. med Manfred Blobner for the opportunity to complete my medical thesis in the department of anesthesiology and intensive care medicine at Klinikum Rechts der Isar.

I am incredibely grateful for the support and guidance Prof. Dr. med. Stefan Schaller and Dr. med. Kristina Fuest provided me over the last few years, starting with the conception and ending with the finalization of my medical thesis. Without their feedback and advice this project would not have been possible.

My thanks also extends to Bernhard Ulm for his statistical support throughout.

Lastly, I would like to thank my parents, especially my father for the immense patience and guidance along the way.

Abstrakt

Einleitung: Eine kritische Erkrankung kann schon früh im Verlauf eines Multiorganversagens Muskelschwund auslösen, der langfristig zu schlechten Behandlungsergebnissen beiträgt. Die Ultraschalluntersuchung des Muskulus (M.) quadriceps femoris kann zur Erkennung und Überwachung von Muskelschwund beitragen. Derzeit fehlen jedoch standardisierte Messprotokolle. Außerdem ist nach wie vor unklar, ob der mit Ultraschall erfasste Muskelschwund mit der Verschlechterung des Funktionsstatus der Patienten zusammenhängt. Das primäre Ziel dieser Beobachtungsstudie war es daher, zu untersuchen, ob die mit Ultraschall gemessene Querschnittsfläche und Dicke des M. quadriceps femoris den Funktionsstatus der Patienten bei der Krankenhausentlassung vorhersagen kann. Sekundäres Ziel war ein Vergleich der Ultraschalltechnik mit dem Goldstandard der CT-Bildgebung zur Quantifizierung des Muskulschwunds bei einer Untergruppe von Patienten.

Methoden: Insgesamt wurden 90 Patienten aus drei prospektiven Beobachtungsstudien an operativen, traumatologischen und neurologischer Intensivpatienten ohne vorherige funktionelle Beeinträchtigung retrospektiv analysiert. Die Querschnittsfläche des M. rectus femoris, die Dicke des M. rectus femoris und die Dicke des M.quadriceps femoris (Summe aus M. rectus femoris plus M. vastus intermedius) wurden bei Aufnahme und Entlassung auf Intensivstation, sowie wöchentlich bei längerem Aufenthalt gemessen. Die funktionelle Abhängigkeit bei Krankenhausentlassung wurde anhand der Mobilitätskomponenten des Barthel-Index gemessen: Transfer (vom Bett zum Stuhl und zurück), Mobilität (auf ebenen Flächen) und Treppensteigen, mit einer maximal erreichbaren Punktzahl von 40 Punkten. Es wurden Spearmans Rangkorrelationskoeffizienten zwischen den Ultraschallmesswerten und den Mobilitätskomponenten des Barthel-Index bei der Krankenhausentlassung bei allen Patienten mit einem Mindestaufenthalt auf Intensivstation von 7 Tagen berechnet. Außerdem wurden Korrelationskoeffizienten zwischen der Gesamtskelettmuskelfläche auf axialen CT-Bildern in Höhe des dritten Lendenwirbels und den Ultraschallmesswerten berechnet.

Ergebnisse: 68 Patienten mit einem mindestens 7-tägigen Aufenthalt auf der Intensivstation wurden in die Analyse einbezogen. 22 Patienten wurden aufgrund von Tod oder Entlassung vor Tag 7 ausgeschlossen. Die mediane Abnahme der Querschnittsfläche des M. rectus femoris bis zur Entlassung aus der Intensivstation betrug 1,69 cm^2 [IQR 2,53 cm^2, 0,53 cm^2]. Die mediane Abnahme der Dicke des M. quadriceps betrug 0,53 cm [IQR 0,74 cm, 0,26 cm], die des M. rectus alleine 0,27 cm [IQR 0,36 cm, 0,01 cm] bis zur Entlassung aus der Intensivstation. Es wurde kein Zusammenhang zwischen der Querschnittsfläche des M. rectus femoris bei der Aufnahme (ρ=-0,01, p=0,918), der Dicke des Rectus femoris (ρ=0,032, p=0,814) oder der Dicke des M. quadriceps (ρ=0,064, p=0,629) und den Werten für den funktionellen Status festgestellt. Außerdem korrelierte die jeweilige Veränderung der Messwerte von der Aufnahme auf der Intensivstation bis zur Entlassung nicht mit der Gesamtveränderung des Funktionsstatus (Querschnittsfläche: ρ=-0,03, p=0,883; Rectus-femoris-Dicke: ρ=-0,12, p=0,510; Quadrizeps-Dicke: ρ=0,08, p=0,630). Bei 22 der 68 Patienten mit CT war die Korrelation zwischen der Skelettmuskelfläche gemessen mit CT und der Querschnittsfläche mit Ultraschall (ρ=0,619; p=0,003) stärker als die mit der Dicke des M. rectus femoris (ρ=0,332; p=0,12) und der Dicke des M. quadrizeps (ρ=0,453; p=0,03).

Schlussfolgerung: Der mit Ultraschall gemessene Muskelschwund war ein schlechter Prädiktor für den Funktionsstatus bei Krankenhausentlassung.

Abstract:
Introduction: Critical illness can trigger muscle atrophy early in the course of multi-organ failure, contributing to poor long-term outcomes. Ultrasound examination of the quadriceps femoris muscle can help detect and monitor muscle atrophy. However, standardised measurement protocols are currently lacking. In addition, it remains unclear whether muscle atrophy detected with ultrasound is related to deterioration in patients' functional status. The primary aim of this observational study was therefore to investigate whether the cross-sectional area and thickness of the quadriceps femoris muscle measured with ultrasound can predict patients' functional status at hospital discharge. The secondary aim was to compare ultrasound technique with the gold standard of CT imaging for quantifying muscle atrophy in a subgroup of patients.
Methods: A total of 90 patients from three prospective observational studies of surgical, trauma and neurological intensive care patients without prior functional impairment were retrospectively analysed. The cross-sectional area of the rectus femoris muscle, the thickness of the rectus femoris muscle and the thickness of the quadriceps femoris muscle (sum of the rectus femoris muscle plus the vastus intermedius muscle) were measured on admission and discharge from the intensive care unit and weekly during longer stays. Functional dependence at hospital discharge was measured using the mobility components of the Barthel Index: Transfer (from bed to chair and back), Mobility (on flat surfaces) and Stair Climbing, with a maximum achievable score of 40 points. Spearman's rank correlation coefficients were calculated between the ultrasound readings and the mobility components of the Barthel index at hospital discharge in all patients with a minimum ICU stay of 7 days. In addition, correlation coefficients were calculated between total skeletal muscle area on axial CT images at the level of the third lumbar vertebra and ultrasound readings.
Results: 68 patients with a minimum 7-day stay in the ICU were included in the analysis. 22 patients were excluded due to death or discharge before day 7. The median decrease in cross-sectional area of the rectus femoris muscle by ICU discharge was 1.69 cm^2 [IQR 2.53 cm^2, 0.53 cm^2]. The median decrease in thickness of the quadriceps muscle was 0.53 cm [IQR 0.74 cm, 0.26 cm], and that of the rectus muscle alone was 0.27 cm [IQR 0.36 cm, 0.01 cm] by ICU discharge. No correlation was found between the cross-sectional area of the rectus femoris at admission (ρ=-0.01, p=0.918), rectus femoris thickness (ρ=0.032, p=0.814) or quadriceps thickness (ρ=0.064, p=0.629) and functional status scores. Furthermore, the respective change in measured values from ICU admission to discharge did not correlate with the overall change in functional status (cross-sectional area: ρ=-0.03, p=0.883; rectus femoris thickness: ρ=-0.12, p=0.510; quadriceps thickness: ρ=0.08, p=0.630). In 22 of the 68 patients with CT, the correlation between skeletal muscle area measured with CT and cross-sectional area with ultrasound (ρ=0.619; p=0.003) was stronger than that with rectus femoris muscle thickness (ρ=0.332; p=0.12) and quadriceps muscle thickness (ρ=0.453; p=0.03).
Conclusion: Muscle atrophy measured by ultrasound was a poor predictor of functional status at hospital discharge.

Table of Contents

Abbreviations 7

List of Figures 8

List of Tables 9

1. Introduction 10

1.1 Background 10

1.2 Skeletal Muscle Ultrasound in ICU Patients 11

1.2.1 Quadriceps Muscle Ultrasound 12

1.2.2 Comparison of Cross-sectional Area and Thickness Measurements 15

1.3 Computer Tomography 17

1.4 Muscle Morphology and Functional Capacity 18

1.5 Goal of this Thesis 19

2. Methods and Materials 20

2.1 Patient Population 20

2.2 Inclusion and Exclusion Criteria 20

2.3 Data Collection 22

2.4 Course of Study 22

2.4.1 Bedside Ultrasound 23

2.4.2 Image Assessment 24

2.4.3 CT Analysis 26

2.5 Statistical Analysis 27

2.5.1 Patient Population 27

2.5.2 Bland-Altman Analysis and Change of Ultrasound Measurement Values 28

2.5.3 Correlation Ultrasound and Functional Capacity 29

2.5.4 Ultrasound and CT Imaging 30

3. Results 31

3.1 Patient Baseline Characteristics 31

3.2 Bland-Altman Plot Analysis 32

3.3 Change in CSA, RF, and QMT from Admission to Discharge 35

3.4 Decline of Muscle Mass on Ultrasound and Functional Capacity 37

3.5 Subgroup Analysis 39

3.6 Comparison of Ultrasound with CT Measurements 43

4. Discussion 46

4.1 Principal Findings **46**

4.2 Comparison with other Studies **46**

4.2.1 Ultrasound 46

4.2.2 Computer Tomography 49

4.3 Strengths and Limitations of Methods **51**

4.3.1 Functional Assessment 51

4.3.2 Ultrasound Methods 52

4.4 Outlook and Future Use **53**

5. Conclusion ***54***

6. References ***55***

7. Appendix ***60***

Abbreviations

APACHE	Acute Physiology and Chronic Health Evaluation
ARDS	Acute respiratory distress syndrome
ASA	American Society of Anesthesiologists
BIA	Bioelectrical impedance analysis
BMI	Body Mass Index
CCI	Charlson Comorbidity Index
CFS	Clinical Frailty Scale
CPR	Cardiopulmonary resuscitation
CSA	Cross-sectional area
CT	Computer tomography
DICOM	Digital Imaging and Communications in Medicine
DXA	Dual-energy x-ray absorptiometry
FIM	Functional Independence Measure
GCS	Glasgow Coma Scale
ICU	Intensive care unit
IQR	Interquartile range
MRC	Medical Research Council
MRI	Magnetic resonance imaging
PACS	Picture Archiving and Communications System
PFIT	Physical Function in Intensive Care Test
QMT	Quadriceps muscle thickness
RF	Rectus femoris
SMA	Skeletal muscle area
SMI	Skeletal Muscle Index
SOFA	Sequential Organ Failure Assessment
VI	Vastus intermedius

List of Figures

Figure 1.1 Anatomy of the quadriceps muscle 13
Figure 1.2 Anatomy of the quadriceps muscle (axial plane) 13
Figure 1.3 Illustration of quadriceps muscle thickness (QMT) measurement 14
Figure 1.4 Ultrasound measuring sites 15
Figure 2.1 Measurement of rectus femoris cross-sectional area (CSA) in cm^2 25
Figure 2.2 Measurement of rectus femoris and quadriceps thickness 25
Figure 2.3 Outline of total skeletal muscle area at the level of the third lumbar vertebra on CT 27
Figure 3.1 Bland-Altman plot analysis of rectus femoris cross-sectional area (CSA) values 33
Figure 3.2 Bland-Altman plot analysis of rectus femoris thickness (RF) values 34
Figure 3.3 Bland-Altman plot analysis of quadriceps muscle thickness (QMT) values 35
Figure 3.4 Plotted change in ultrasound measurement values and Mobility-Transfer-Barthel-Index 38
Figure 3.5 Change in rectus femoris cross-sectional area by degree of decline in functional capacity 40
Figure 3.6 Change in rectus femoris thickness by degree of decline in functional capacity 41
Figure 3.7 Change in quadriceps muscle thickness by degree of decline in functional capacity 42
Figure 3.8 Correlation between CT skeletal muscle area (cm^2) and ultrasound measurement values 44

List of Tables

Table 2.1 Inclusion and exclusion criteria 21
Table 3.1 Patient characteristics 32
Table 3.2 Absolute change in ultrasound measurements presented as median [IQR] 36
Table 3.3 Percentage change of ultrasound measurements presented as median [IQR] 37
Table 3.4 Correlation between ultrasound measurement values and Mobility-Transfer-Barthel-Index 39
Table 3.5 Correlation between changes in ultrasound values and Mobility-Transfer-Barthel-Index 39
Table 3.6 Change on ultrasound by complete vs. incomplete dependency 43
Table 3.7 Ultrasound measurement values stratified by sarcopenia on CT 45
Table 4.1 Comparison of results with similar studies 47

1. Introduction

1.1 Background

Critical illness and admission to an intensive care unit represents a burden on patients, that reaches beyond a short-term acute decline in health, and can have a lasting impact on long-term physical and psychological outcomes in survivors well beyond discharge. Survivors of critical illness often experience impairment in the ability to perform activities of daily living, cognitive decline, reduction in exercise tolerance, challenges recovering to their pre-illness status, and reduced quality of life (Iwashyna, Ely et al. 2010, Desai, Law et al. 2011). The often experienced psychological and emotional burden of critical illness associated with depression, anxiety and posttraumatic stress disorder (PTSD), decline in memory and concentration, and loss of functional independence and mobility can be summarized as the post intensive care syndrome (PICS) (Rawal, Yadav et al. 2017). These challenges have shown to persist not only in the weeks and months following discharge from the ICU, but have afflicted patients up to five years following critical illness (Herridge, Tansey et al. 2011). Certain ICU populations such as those surviving severe acute respiratory distress syndrome (ARDS), sepsis, trauma, or prolonged mechanical ventilation tend to be particularly affected by poor quality of life following discharge (Oeyen, Vandijck et al. 2010). Alongside the life-threatening illnesses warranting ICU admission, factors such as immobilization, inflammation, and oxidative stress lead to a state of muscular atrophy dominated by increased protein breakdown and decreased protein synthesis (Batt, Herridge et al. 2019). This can contribute to the development of critical illness myopathy (CIM) and intensive care unit-acquired weakness (ICUAW). ICUAW is a syndrome of clinically apparent weakness not explained by other etiologies other than critical illness (Lad, Saumur et al. 2020). Although the development of weakness in critically ill patients is a common issue in the ICU, muscular atrophy does not entirely explain this clinical syndrome (Lad, Saumur et al. 2020). Other risk factors such as the severity of illness, sepsis, multiorgan failure, hyperglycemia and the use of corticosteroids and neuromuscular blocking agents have been associated with the development of ICUAW (Kress and Hall 2014). ICUAW is associated with prolonged mechanical ventilation (De Jonghe, Sharshar et al. 2002), longer hospital stay, and increased mortality (Ali, O'Brien et al. 2008). This syndrome remains mainly a clinical diagnosis, that can be supported by muscle biopsies and

electrophysiological studies. Lastly, the quantification of skeletal muscle remains difficult and is influenced by many factors. The role of imaging, specifically of ultrasound and its potential as a tool to monitor skeletal muscle mass and support the diagnosis of weakness remains unclear.

1.2 Skeletal Muscle Ultrasound in ICU Patients

Ultrasound as a tool to assess muscular status and identify muscle wasting in critically ill patients has been investigated with growing interest in the past decade. Skeletal muscle wasting has been shown to occur early in the course of critical illness. Thus, a tool that can reliably detect changes early into ICU stay and identify patients at most risk for muscle loss is needed (Puthucheary, Rawal et al. 2013). Yet, it is unclear which diagnostic tool is best suited to aid clinicians with this task. While there are many techniques to quantify skeletal muscle mass, such as bioelectrical impedance analysis (BIA), dual energy x-ray absorptiometry (DXA), computer tomography (CT), and magnetic resonance imaging (MRI), not all modalities are feasible in a critically ill population (Guerri, Mercatelli et al. 2018, Looijaard, Molinger et al. 2018). Ultrasound however, is a non-invasive and reproducible diagnostic tool, that is readily available at the bedside, quick to perform, and requires no great expertise. It is thus a feasible tool to perform serial measurements and monitor skeletal muscle during ICU stay (Thomaes, Thomis et al. 2012, Tillquist, Kutsogiannis et al. 2014, Mueller, Murthy et al. 2016). Additionally, there is no patient cooperation required for the assessment of skeletal muscle with ultrasound, avoiding the frequent issue of patient sedation hindering clinical evaluation. Given these advantages, strict adherence to an ultrasound measuring protocol is essential to establish reproducible and comparable results (Mourtzakis, Parry et al. 2017). Unfortunately, the standardization of ultrasound measuring protocols in clinical research is lacking, making direct comparisons between studies difficult (Ticinesi, Meschi et al. 2017). Factors while measuring, such as the positioning of the patient in bed, the selection of the ultrasound probe, the anatomical landmarks used to find measurement sites on the skin, the angle at which the probe is held while measuring, the amount of compression applied, as well as the image evaluation procedure, all influence reproducibility of ultrasound data (Fischer, Anwar et al. 2020).

Another question is, which muscle best serves to represent muscle wasting in immobilized critically ill patients. Regional differences of age-related muscle loss measured by ultrasound have

been shown to exist throughout the body. For instance, Abe et al., using ultrasound in a healthy Japanese population, demonstrated that age-related loss of upper-thigh muscle can be detected sooner than that of whole-body muscle (Abe, Thiebaud et al. 2014). Additionally, the authors found the prevalence of age-related thigh muscle loss to be higher than that of other measuring sites such as the upper-arm or trunk (Abe, Loenneke et al. 2014). Even differences in the rate of muscle wasting amongst different lower limb muscles have been observed. Annetta et al. found a significant 45% decrease in rectus femoris (upper leg muscle) cross-sectional area (CSA) over a 20-day ICU stay in a cohort of 38 young trauma patients while the tibialis anterior (lower leg muscle) CSA reduction of 22% was non-significant (Annetta, Pittiruti et al. 2017). The authors discussed whether different functions and muscle fiber types played a role in the different magnitudes of change for upper and lower leg muscles. As the authors state, the rectus femoris is an extensor with type II fast fiber predominance and the tibialis anterior a flexor with type I slow fiber predominance. The mechanism responsible for these differences in reduction rates amongst different muscle groups remains unclear (Annetta, Pittiruti et al. 2017). Certainly, the accessibility of the ultrasound measurement site plays a role in deciding how many and which muscles to assess in a critically ill patient. Studies training novice operators to perform quadriceps ultrasound have shown quick proficiency with high inter-observer reliability (Mueller, Murthy et al. 2016).

1.2.1 Quadriceps Muscle Ultrasound

Ultrasound assessment of the quadriceps, a four bodied muscle comprised of the rectus femoris, vastus medialis, vastus lateralis, and vastus intermedius muscles, has been used extensively to assess muscle wasting in the critically ill (Weinel, Summers et al. 2019)(see Figures 1.1-2). Specifically, two common measurements are used: muscle cross-sectional area (CSA) and muscle thickness. The rectus femoris, an anterior thigh muscle required for flexion in the hip joint, is often used to evaluate muscle CSA as a surrogate for whole-body skeletal muscle (van Ruijven, Stapel et al. 2021). For muscle thickness, either the rectus femoris or quadriceps (sum of rectus femoris and vastus intermedius muscle thickness) is frequently measured (see Figure 1.3). For the purpose of this thesis, CSA will refer to the cross-sectional area and RF to the thickness of the rectus femoris muscle. QMT will refer to the total thickness of the quadriceps muscle.

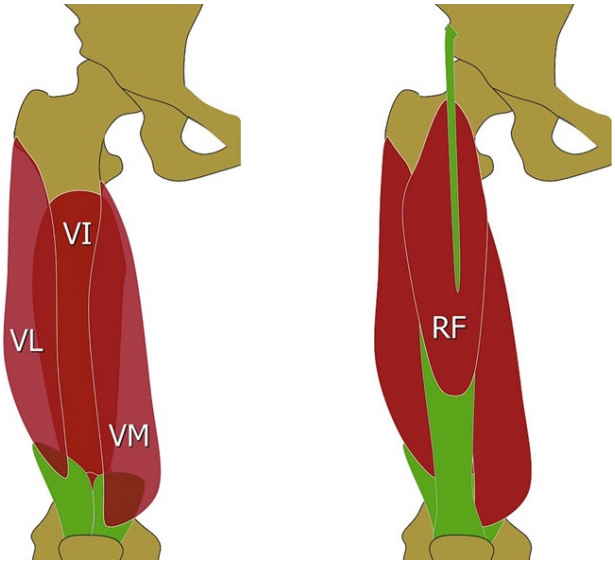

Figure 1.1 Anatomy of the quadriceps muscle. Left image = deep plane; right image = superficial plane; VL = vastus lateralis; VI = vastus intermedius; VM = vastus medialis; RF = rectus femoris. Modified after "Sonography of the quadriceps muscle: Examination technique, normal anatomy, and traumatic lesions" (Pasta, Nanni et al. 2010).

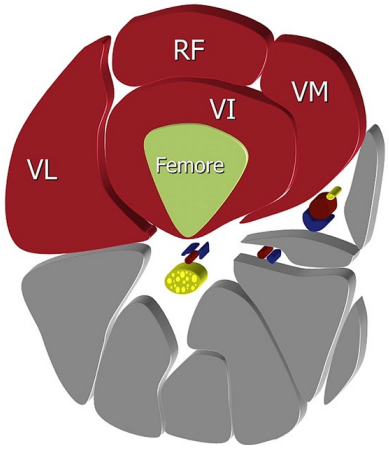

Figure 1.2 Anatomy of the quadriceps muscle (axial plane). VL = vastus lateralis; VI = vastus intermedius; VM = vastus medialis; RF = rectus femoris. Modified after "Sonography of the quadriceps muscle: Examination technique, normal anatomy, and traumatic lesions" (Pasta, Nanni et al. 2010).

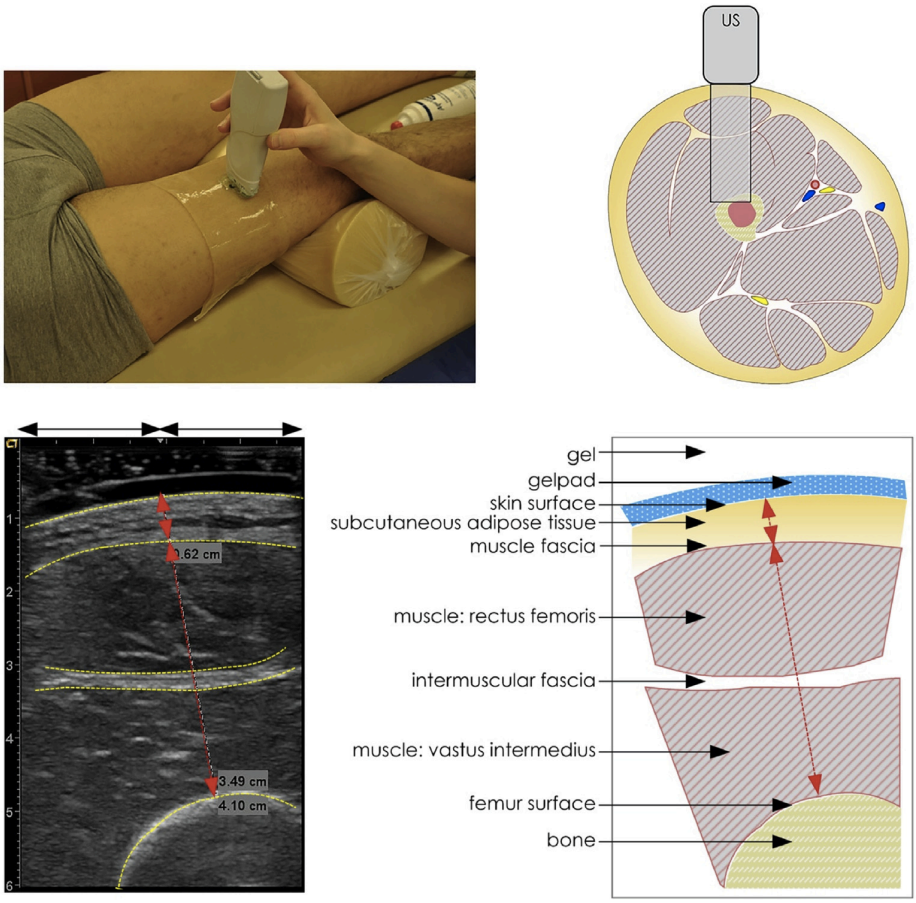

Figure 1.3 Illustration of quadriceps muscle thickness (QMT) measurement. QMT: sum of the rectus femoris and vastus intermedius thickness, via ultrasound at the anterior mid-thigh (long red arrow). Subcutaneous adipose tissue thickness is shown by the short red arrow. Modified after "Ultrasound method of the USVALID study to measure subcutaneous adipose tissue and muscle thickness on the thigh and upper arm: An illustrated step-by-step guide" by Fischer, A., Anwar, M., Hertwig, A., Hahn, R., Pesta, M., Timmermann, I., Siebenrock, T., Liebau, K., and Hiesmayr, M., 2020, Clinical Nutrition Experimental, 32: 38-73.

As illustrated in Figure 1.4, measurements are commonly performed at the mid-thigh, with reports of measurement sites at 1/2, 2/3, or 3/5 distance from the anterior superior iliac spine of the pelvis to the upper border of the patella throughout the literature (Weinel, Summers et al. 2019).

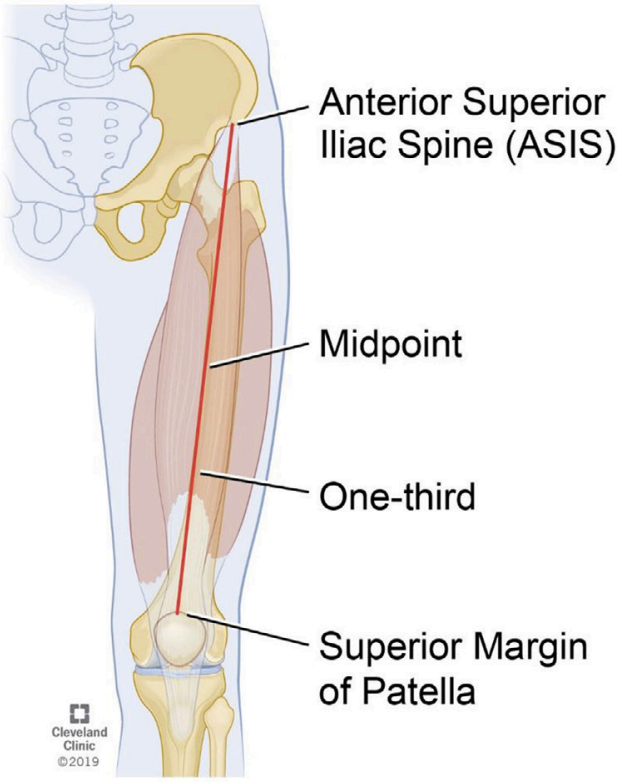

Figure 1.4 Ultrasound measuring sites at 1/2 (Midpoint) and 2/3 (1 – 1/3; One-third) distance from the anterior superior iliac spine (ASIS) to the upper border of the patella. (Bury, Dechicco et al. 2021).

1.2.2 Comparison of Cross-sectional Area and Thickness Measurements

Although there has been extensive research into these two individual measurements, it remains unknown whether the CSA or thickness measurement is most suitable to assess muscular status and predict functional disability in critically ill patients. Though many studies have measured change in muscle mass over time, fewer have explored the relationship between these measurements and clinical outcomes such as strength or functional capacity.

Katari et al. showed a highly significant loss of rectus femoris and vastus intermedius thickness from day 1 to day 7 in 100 patients of a multidisciplinary ICU (Katari, Srinivasan et al. 2018). Baldwin compared the thickness and physical strength of multiple muscles in 16 patients with sepsis to 16 healthy individuals. The authors found that the quadriceps had significantly greater thickness decline than the arm muscles in the ICU patients compared to control subjects after a median of 16 days (Baldwin CE 2014). Similarly, Hadda et al. performed serial thickness measurements of the quadriceps on days 1, 3, 5, and 7 in 70 ICU patients with sepsis. The authors

found a significant decline of 10% after one week. In addition, there was greater muscle thickness in survivors compared to non-survivors at all measurement times. A significantly higher thickness decline was associated with poor 90-day outcome, which was defined as unplanned hospital readmission (Hadda, Kumar et al. 2018).

Serial measurements of the CSA in ICU patients have also shown significant muscle decline over ICU stay. Not many studies have prospectively compared histopathological muscle biopsies, a gold standard for skeletal muscle wasting, with serial ultrasound measurements. In a study with 63 ICU patients, Puthucheary et al. found a significant reduction in rectus femoris CSA on ultrasound (-10.5%) and vastus lateral CSA on biopsy (-12.5%) over the first 7 days of ICU stay (Puthucheary, Rawal et al. 2013).

Few studies have compared the change in muscle CSA head-to-head with the change in quadriceps muscle thickness (QMT) on ultrasound over the course of ICU stay and the relationship between these changes and functional capacity. One study conducted by Palakshappa et al. prospectively measured rectus femoris CSA and QMT on day 1 and day 7 of ICU stay with ultrasound in 29 patients with sepsis. The authors assessed the patients' muscle strength on day 7 using the Medical Research Council (MRC) sum score (Kleyweg, Van Der Meché et al. 1991). Patients' functional capacity (ability to perform activities of daily living) was measured on day 7 using the Physical Function ICU Test (PFIT) (Skinner, Berney et al. 2009). While a 23.2% reduction in rectus femoris CSA over 7 days correlated moderately with strength on MRC score (ρ 0.51, p=0.03), a QMT decrease of 17.9% did not correlate with either strength (ρ −0.07, p=0.77) or functional capacity (ρ −0.11, p=0.68) (Palakshappa, Reilly et al. 2018). Similar to Palakshappa et al., Puthucheary et al. advocate for the use of rectus femoris CSA over muscle thickness measurements based on results of another study comparing ultrasound with biopsy (Puthucheary, McNelly et al. 2017). The authors compared both ultrasound methods with a histopathological (myofiber CSA) and biochemical (Protein: DNA ratio) result on biopsy over 7 days in 19 mechanically ventilated patients. The decline in QMT measured on ultrasound significantly underestimated the decline in myofiber CSA (- 4.6% vs. - 16.4%; p=0.025) and protein: DNA ratio (- 4.6% vs. - 30.9%; p=0.019). They also found no difference in QMT change after one week between those with and without knee extension weakness (12.6% vs. 12.1%, respectively; p=0.95), measured by the MRC score on day 10. Lastly, there was a significant difference in CSA change between weak and non-weak patients (20.7% vs. 8.4%, respectively; p=0.012) (Puthucheary,

McNelly et al. 2017), strengthening their case that CSA may be the preferable measurement over thickness. In conclusion, both CSA and thickness measurements have shown muscle mass decline early during the course of critical illness. Additionally, impairment of strength and physical function has been inconsistently associated with either single time muscle measurements or serial measurements of muscle decline. It thus remains unclear which ultrasound measurement correlates best with muscle loss and functional decline.

1.3 Computer Tomography

Another imaging technique that plays an indispensable role in the diagnostic evaluation of patients is computer tomography (CT). The role of CT has also been firmly established as a gold standard method in body composition analysis, that reliably quantifies skeletal muscle mass and visceral adipose tissue (Guerri, Mercatelli et al. (2018). Further, single slice axial CT measurements of skeletal muscle are strongly correlated with whole-body CT measurements (r = 0.71- 0.92) (Shen, Punyanitya et al. 2004). Commonly, the height of the third lumbar vertebra on CT imaging is the site for quantitative analysis of these tissues. Often, the term sarcopenia is used to describe patients below established cut-off values for skeletal muscle mass. The European Working Group on Sarcopenia in Older People's (EWGSOP) 2018 definition defines sarcopenia as a generalized skeletal muscle disease associated with impaired muscle strength and low muscle quantity or quality (Cruz-Jentoft, Bahat et al. 2019). Briefly, these sarcopenia cut-off values are calculated by taking the total area of the following muscles at the third lumbar vertebra from a single slice CT image: rectus abdominis, internal and external obliques, transversus abdominis, psoas, quadratus lumborum, and erector spinae (see Figure 2.3). This area is then adjusted to the square height of the patient, yielding a skeletal muscle index (cm^2 /m^2) (Mourtzakis, Prado et al. 2008). Different software programs applied for body composition analysis, such as ImageJ from the National Institute of Health (Bethesda, MD, USA) and SliceOmatic (Tomovision, Magog, Canada), have shown excellent interrater reliability between programs (Teigen, Kuchnia et al. 2018). Further, research using CT for body composition analysis has been conducted in certain patient populations (cancer, advanced liver disease, and critically ill) demonstrating a consistent correlation between low muscle mass and adverse outcomes (Prado, Lieffers et al. 2008, Englesbe, Patel et al. 2010, Sheean, Peterson et al. 2014). For instance, Moisey et al. analyzed the abdominal CT scans of 149

elderly trauma patients admitted to the ICU. They divided their cohort into sarcopenic and non-sarcopenic patients based on established skeletal muscle index cut-off points. The authors found that those patients with sarcopenia had fewer ventilator-free days, ICU-free days, and higher mortality compared to those without (Moisey, Mourtzakis et al. 2013). Similarly, Ng et al. retrospectively analyzed 228 mainly surgical ICU patients with CT scans performed within 72 hours of admission. Patients with low muscularity had increased odds of hospital mortality (Odds Ratio 2.4), independent of age, admission category, disease severity, or number of failing organs (Ng, Lee et al. 2020). Interestingly, Loosen et al. followed critically ill patients over 6 and 12 months, finding that survivors showed significantly greater skeletal muscle mass on admission CTs compared with non-survivors (Loosen, Schulze-Hagen et al. 2020). Although reliable, CT imaging has disadvantages when compared to other modalities: for instance, the expertise required to analyze images as well as specific software requirements for body composition hinder a broad clinical application to monitor skeletal muscle mass (Joskova, Patkova et al. 2018). Additionally, the exposure to ionizing radiation poses a risk to the patient that modalities such as ultrasound do not. This limits the use of CT for serial assessments to detect changes in body composition over time. Specifically for the critically ill population, the risks associated with long transport times from the ICU to the CT scanner do not warrant assessment solely to quantify skeletal muscle (Looijaard, Molinger et al. 2018). Lastly, the high costs associated with CT imaging has limited this technique mainly to the research setting or to populations in which imaging is part of routine care (Heymsfield, Gonzalez et al. 2015). In conclusion, CT has proven to be an accurate and reliable tool to quantify skeletal muscle using specialized software in different patient populations.

1.4 Muscle Morphology and Functional Capacity

The quadriceps muscle is easily accessible for assessment with ultrasound and its size has repeatedly been correlated with strength in a number of populations, including the critically ill (Mourtzakis, Parry et al. 2017). Although clinical strength tests such as the MRC sum score are recommended to diagnose weakness in critically ill patients, they are limited by the requirement of a fully awake and cooperative patient (Hermans and Van den Berghe 2015). Arguably of greater clinical relevance is the decrease in functional capacity which patients experience after ICU stay. Poor functional capacity at hospital discharge has been associated with increased 90-day post-

discharge mortality (Rydingsward, Horkan et al. 2016). The question remains if there is a relationship between quadriceps morphology and functional capacity following ICU and hospital discharge. Another question is which ultrasound measurement, CSA or thickness, best predicts functional capacity at discharge. The benefits of both measurements are rarely compared in studies (Weinel, Summers et al. 2019). Further, inconsistencies amongst studies in measuring protocols and the uncertainty about measurement frequency to monitor changes complicate comparison of these studies (van Ruijven, Stapel et al. 2021). The lack of standardized cut-off values to define low muscle mass on ultrasound makes it challenging to classify patients who truly have low muscle mass (Perkisas, Baudry et al. 2018). In addition, stratifying patients by muscle mass to identify those at highest risk of complications associated with muscle wasting is challenging (Wilkinson, Gore et al. 2021). In order to advance the understanding of whether changes in muscle morphology on ultrasound translate to changes in functional capacity, further comparisons of ultrasound measurements with functional outcomes are needed.

1.5 Goal of this Thesis

The goal of this thesis is to explore the relationship between two different ultrasound measurements of muscle mass and mobility at discharge in critically ill patients with a minimum stay of 7 days in the ICU. We hypothesized that the loss of muscle mass over ICU stay could predict a patient's decline in functional independence on transfer from bed to chair and mobility on level surfaces. Additionally, we aimed to compare ultrasound with a CT imaging technique to quantify skeletal muscle in the subset of patients with abdominal scans on ICU admission.

The following assessments were performed:

1. Comparison of the rectus femoris CSA measurement with the rectus femoris and quadriceps thickness measurement using ultrasound in relation to functional independence in activities of daily living at hospital discharge, as assessed by the components "mobility" and "transfer" of the Barthel-Index.
2. Comparison of CSA and thickness measurements on ultrasound with total CSA of skeletal muscle on lumbar CT scan.

2. Methods and Materials

2.1 Patient Population

Between March 2017 and April 2020, we collected data in a prospective database from 90 critically ill patients who were admitted to the intensive care unit at Klinikum Rechts der Isar university hospital. This cohort of patients was comprised of three study populations each with serial ultrasound assessments: trauma patients included in the Miracle II Study (Metabolomics pilot study on intensive care acquired muscle weakness), stroke patients from the NICU study (Effects of mobility dose on discharge disposition in critically ill stroke patients), and general surgical ICU patients from the SICU-SOMS2 study (Effects of mobility dose in surgical intensive care unit patients on muscle wasting and adverse outcomes). For the purpose of this thesis, only patients with a minimum stay of 7 days in the ICU were included for analysis.

2.2 Inclusion and Exclusion Criteria

Trial inclusion and exclusion criteria are provided in Table 2.1. Age requirement for all three trial populations was 18 years or over. Informed consent was obtained from patients directly, and if not possible, from legal proxy.

Table 2.1 Inclusion and exclusion criteria		
Study	**Inclusion criteria**	**Exclusion criteria**
Miracle II	• Trauma in the 24 hours prior to inclusion • Pre-operative physical function score of ASA I or II prior to admission (healthy or mild systemic disease) • Expected to remain in the ICU for no less than three days	• CPR performed following trauma prior to admission • Therapy strategy deescalated to comfort care
NICU	• Ischemic stroke or non-traumatic intracerebral hemorrhage • Admission to the ICU no longer than 48 hours prior to inclusion • Functional independence exhibited by a Barthel-Index > 70 points two weeks prior to hospital admission	• Transfer from another institution (other hospital, rehabilitation center, skilled nursing facility, etc.) • Exclusive or clinically dominant posterior circulation stroke, subarachnoid hemorrhage, subdural or epidural hemorrhage • Absence of lower limbs • Therapy strategy deescalated to comfort care
SICUSOMS2	• Functional independence exhibited by a Barthel-Index > 70 points prior to hospital admission • Admission to the ICU no longer than 48 hours prior to inclusion • Expected to remain in the ICU for no less than three days	• Transfer from another institution • Absence of lower limbs • Pregnancy • Therapy strategy deescalated to comfort care • High risk for persistent brain injury (GCS <5), ischemic stroke, or non-traumatic intracranial hemorrhage
ASA American Society of Anesthesiology *CPR* Cardiopulmonary resuscitation *GCS* Glasgow Coma Scale		

2.3 Data Collection

Data obtained upon admission were age, gender, body mass index (BMI), location prior to ICU admission (home or other hospital), reason for ICU admission, medical department of care at admission, Barthel-Index two weeks prior to hospital admission, admission Glasgow Coma Scale (GCS), Charlson Comorbidity Index (CCI), Clinical Frailty Scale (CFS), admission Acute Physiology and Chronic Health Evaluation II Score (APACHE II), admission Sequential Organ Failure Assessment (SOFA) score, as well as standard laboratory and hemodynamic parameters.

The GCS assesses a patient's level of consciousness (eye movement, verbal, and motor response to stimulus), ranging from 3 to 15 points representing coma to full alertness, respectively (Teasdale and Jennett 1974). The Charlson Comorbidity Index (CCI) is used to predict 10-year mortality by weight of certain comorbidities (Charlson ME 1987). The Clinical Frailty Scale (CFS) assesses aspects such as a patient's cognition, function, and overall dependence in activities of daily living and ranges from 1 (very fit) to 9 (terminally ill) (Rockwood, Song et al. 2005). The level of frailty two weeks prior to hospital admission was obtained for the purpose of this database. The Apache II Score is used to predict ICU mortality based on a patient's acute physiological derangement (Acute Physiology Score), age (Age Points), and previous health status (Chronic Health Points). A maximum possible score of 71 points indicates highest risk of death (Knaus WA 1985). The SOFA score is used to quantify a patient's level of organ dysfunction and ranges from 0-4 for six different organ systems (Vincent JL 1996). All scoring tables are listed in the appendix.

2.4 Course of Study

For all patients, ultrasound measurements were performed within 48 hours of admission to the ICU and were repeated weekly or within one day of ICU discharge. Upon ICU discharge, data were obtained regarding ICU length of stay and functional mobility status, assessed by the components "transfer", "mobility" and "stairs" of the Barthel-Index. The Barthel-Index is a 100-point test assessing the degree of functional independence and required assistance for certain activities of daily living (see Appendix). For the purpose of the prospective database, a score for the following three Barthel-Index components was obtained: 1. Transfer (from bed to chair and back) 2. Mobility

on level surfaces and 3. Stairs (ability to climb) with a maximum achievable score of 40 points. The lowest score represented complete functional dependence and the highest score complete functional independence. Scoring was obtained via direct questioning of nursing staff, physical therapists, or treating physicians. Upon hospital discharge, data on hospital length of stay, discharge disposition (discharge to prior residence, nursing home, rehabilitation clinic, etc.) and functional independence, again measured through three components of the Barthel-Index (maximum score of 40 points), were obtained. For the primary endpoint analysis, the components "transfer" and "mobility" of the Barthel-Index were used. Subsequently, these components will be referred to as the Mobility-Transfer-Barthel.

2.4.1 Bedside Ultrasound

Within 48 hours of ICU admission, ultrasound image acquisition of the thigh muscle was performed using either a 2-6 MHz curvilinear probe on the Sparq Ultrasound System (Philips, Bothell, WA, USA) or a 1.8-5 MHz 4C-SC curvilinear probe on a GE ultrasound device (GE Healthcare, Chicago, Illinois, USA). Prior to assessment, patients were placed into the supine position with their upper body raised to a 30° angle and their legs relaxed and extended outright in bed. First, the CSA and thickness measuring sites were identified by marking 3/5 of the distance from the superior anterior iliac spine (ASIS) to the mid-upper border of the patella with a pen (Mueller, Murthy et al. 2016). Except for the CSA measurement in the SICUSOMS2 patients (2/3 distance), CSA and thickness measurements were all taken at 3/5 distance (Parry, El-Ansary et al. 2015). After applying ultrasound gel, the probe was placed at a 90° angle perpendicular to the longitudinal axis of the thigh. Two measurements, one to acquire the CSA of the rectus femoris muscle (CSA in cm^2), and the other to obtain the thickness of the rectus femoris and quadriceps muscle (RF and QMT in cm) were performed to yield three values per leg. For the SICUSOMS2 cohort, the right leg was used as the measurement standard. The same limb was then serially measured at each measuring time point. The measurement of the CSA was acquired by applying minimal pressure with the probe. Images ideally captured a centered muscle with visualization of the femur bone. As previously described, muscle thickness images were captured with maximum compression on the ultrasound probe (Paris, Mourtzakis et al. 2017). All images were saved and exported to a USB device for later assessment.

2.4.2 Image Assessment

All Images were imported into HOROS, a free, open-source medical image viewing software (LGPL-3.0; Lesser General Public License, Version 3.3.6) and analyzed by a single assessor (HL). Image quality control for all 90 patients was performed prior to assessment to ensure the muscle was centered and that the left and right borders as well as the hyperechoic fascia encircling the rectus femoris muscle were visible. Images which did not meet these criteria were excluded from assessment. Similarly, quality of muscle thickness images was assessed, ensuring the rectus femoris and vastus intermedius muscles were centered underneath the probe, and not squeezed to one side during maximum compression. Images that did not meet these criteria were excluded from assessment. First, with the use of a free hand tracing tool, the CSA was marked along the hyperechoic fascia of the rectus femoris muscle (see Figure 2.1). Next, the thickness of the rectus femoris and quadriceps muscle (rectus femoris and vastus intermedius in cm) was measured along the shortest imaginary line from the exact top-center of the image, down to the muscle-bone interface (Fischer, Anwar et al. 2020). One distance was measured along this imaginary line (rectus femoris), before a second line was placed over the quadriceps to yield two thickness values (see Figure 2.2).

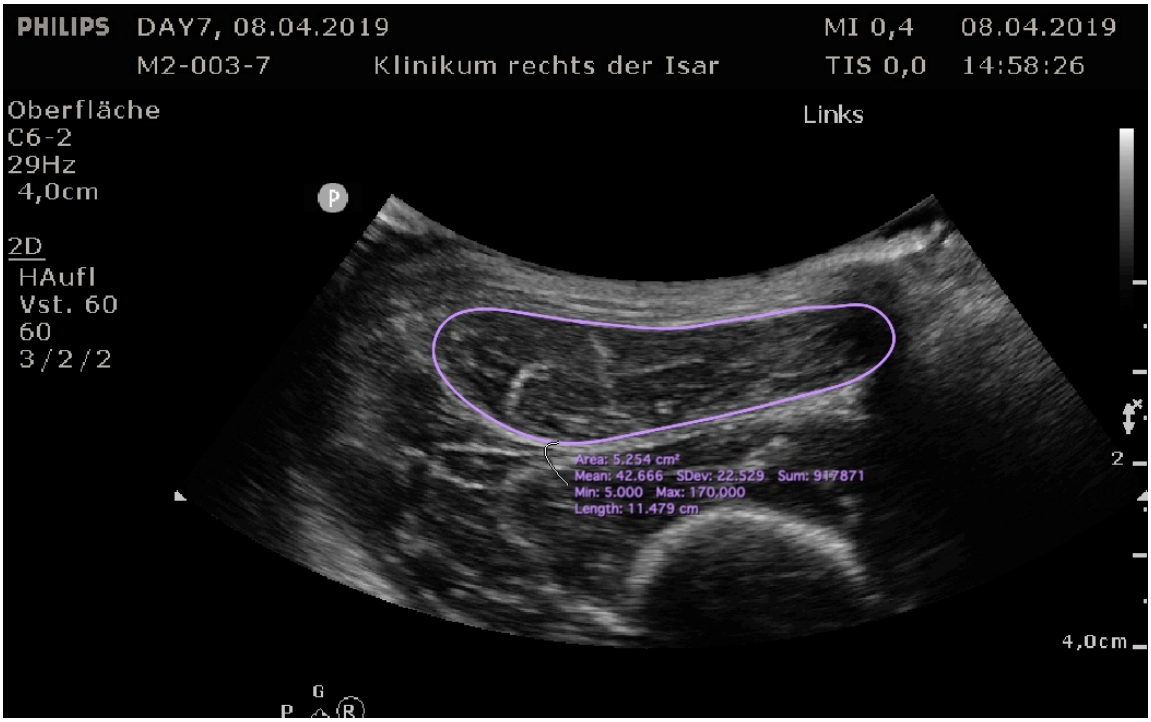

Figure 2.1 Measurement of rectus femoris cross-sectional area (CSA) in cm^2.

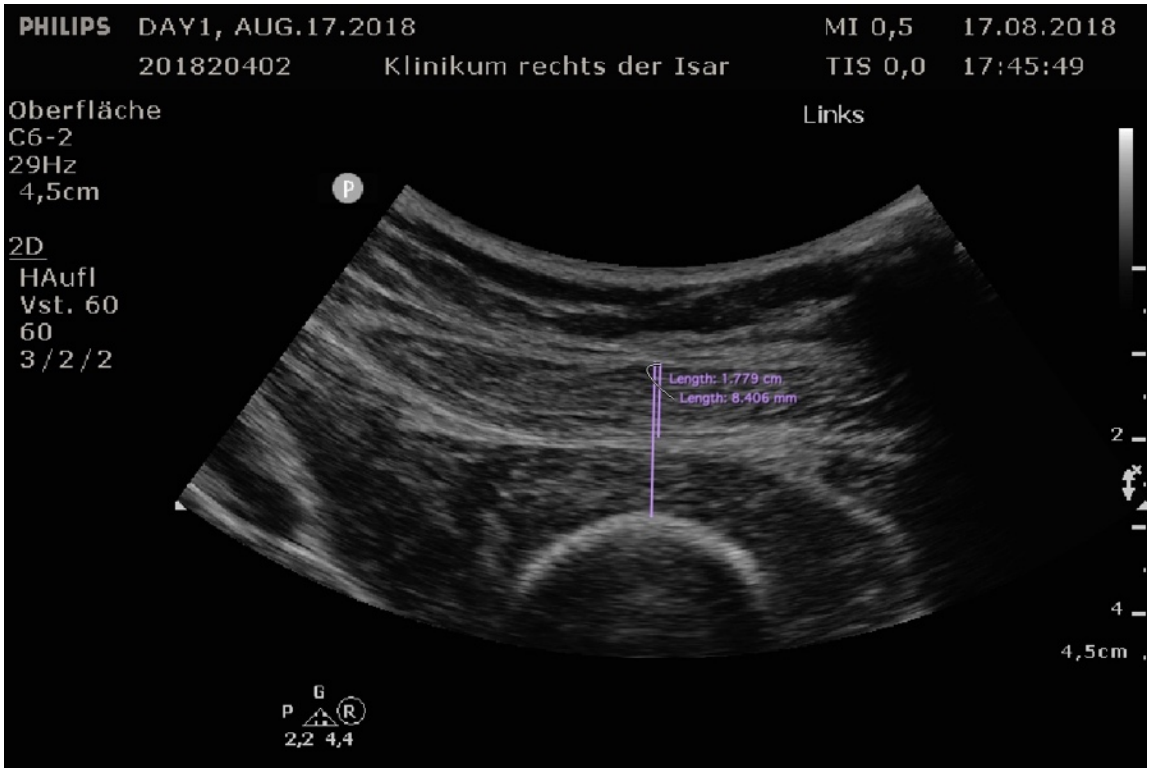

Figure 2.2 Measurement of rectus femoris (0.84 cm) and quadriceps thickness (1.78 cm).

2.4.3 CT Analysis

All patients were screened retrospectively for having one or more performed CT scans that included the third lumbar vertebra during ICU or hospital stay. First, images were viewed on an internal hospital picture archiving and communications system (PACS) by a single assessor (HL). Sagittal axis view of the vertebrae was used to simplify identifying the height of the third lumbar vertebra. Next, the midpoint on sagittal axis view of the third lumbar vertebra was marked and the axial slice image at this anatomical point was saved and exported in a DICOM format to an external USB device. Once exported, images were analyzed using a predefined protocol for body composition analysis with ImageJ software version 1.53 from the National Institute of Health (Bethesda, Maryland, USA) (Gomez-Perez, Haus et al. 2016). Prior to analysis, images were assessed according to protocol for quality control, ensuring the circumference of the abdominal wall was fully visible and no metal artifacts that could interfere with thresholding were present (Gomez-Perez, Haus et al. 2016). Images that did not meet these criteria were excluded from assessment. First, three areas outlined by the following layers were traced on the image using the "Freehand selection" tool: outer abdominal muscle layer, inner abdominal muscle layer, and area of the vertebral head (see Figure 2.3). Next, a predefined attenuation threshold of –29 to 150 Hounsfield Units, specific for skeletal muscle, was set to calculate the skeletal muscle area (cm^2) on the axial slice image (Mourtzakis, Prado et al. 2008). In order to calculate the skeletal muscle area, the latter two areas were subtracted from the former and thus included the following muscles: rectus abdominus, internal and external obliques, transverse abdominus, erector spinae, quadratus lumborum and psoas. Then, to convert from mm^2 to cm^2, the skeletal muscle area value was divided by 100. Further, the skeletal muscle index (SMI) to define low muscle mass was derived by dividing the skeletal muscle area by the square height of the patient (cm^2/m^2). Finally, according to well established cut-off values, a skeletal muscle index below 52.4 and 38.5 was deemed to define sarcopenia for male and female patients, respectively (Prado, Lieffers et al. 2008).

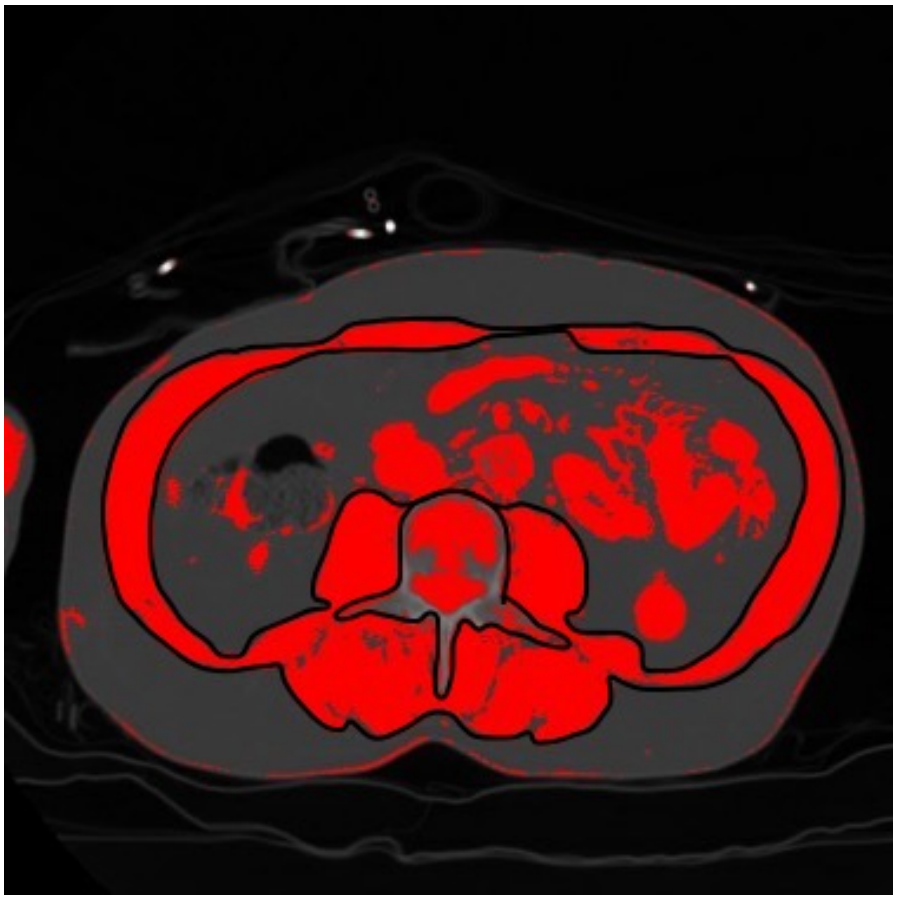

Figure 2.3 Outline of total skeletal muscle area at the level of the third lumbar vertebra on CT imaging using ImageJ. To calculate the total skeletal muscle area, the area outlining the vertebral head and inner abdominal muscle layer was subtracted from the area outlined by the outer abdominal muscle layer.

2.5 Statistical Analysis

2.5.1 Patient Population

Of 90 patients admitted to the ICU with ultrasound assessment upon admission, 68 patients with a minimum stay of 7 days were included for statistical analysis. 22 Patients were excluded because of death or discharge from the ICU prior to day 7. After image quality control using the afore mentioned criteria was performed, all measurement values were recorded in a Microsoft Excel spreadsheet. Images that did not meet the criteria of quality control were excluded. Ultrasound assessments that could not be performed were marked as missing. In those patients with bilateral ultrasound assessment and an inadequate image on one side, the measurement value of the other leg was used for all calculations. All statistical analyses were performed using the statistics

software R version 4.1.0 (2021-05-18). Descriptive statistics are reported as median with interquartile ranges (IQR) and categorical variables as frequencies and percentages.

2.5.2 Bland-Altman Analysis and Change of Ultrasound Measurement Values

In all patients with bilateral ultrasound, Bland-Altman plot analysis was first performed to assess agreement and detect potential measurement bias between the left and right leg. This was repeated for each separate method of measurement: rectus femoris CSA, rectus femoris thickness (RF), and quadriceps thickness (QMT). Limits of agreement on the Bland-Altman plots are defined as the limits within which 95% of the difference between left and right measurement values lies.

The maximum, minimum, and mean ultrasound values at admission and ICU discharge were each used in patients with both left and right leg values to test if this selection influenced absolute change over time. Calculation of absolute change was repeated for all three measurement methods: CSA, RF, and QMT. Only patients with an ultrasound assessment on the day of ICU discharge were included. Patients with inadequate or missing images on ICU admission or discharge were excluded from calculation. The following formula was used with the maximum, minimum, and mean values of CSA, RF, and QMT:

Formula to calculate absolute change of ultrasound measurement values

$$\text{Absolute Change } CSA_{max} = CSA_{max} \text{ ICU Discharge } (cm^2) - CSA_{max} \text{ ICU Admission } (cm^2)$$

$$\text{Absolute Change } CSA_{mean} = CSA_{mean} \text{ ICU Discharge } (cm^2) - CSA_{mean} \text{ ICU Admission } (cm^2)$$

$$\text{Absolute Change } CSA_{min} = CSA_{min} \text{ ICU Discharge } (cm^2) - CSA_{min} \text{ ICU Admission } (cm^2)$$

In order to strengthen a comparison between CSA and thickness, percentage change from admission to Day 7 and ICU discharge across all measured images for CSA, RF, and QMT was calculated, irrespective of image quality. If ultrasound was not available on the day of ICU discharge, the percentage change to the last ultrasound assessment in the ICU was calculated. Therefore, a calculation was possible in 65 of the 68 patients with a minimum stay of 7 days in the ICU. The following formula was used:

Formula to calculate percentage change of ultrasound measurement values

% Change CSA Day 1 to Day 7 = (CSA Value day 7 – CSA Value Day 1) / CSA Value Day 1 * 100

% Change RF Day 1 to Day 7 = (RF Value Day 7 – RF Value Day 1) / RF Value Day 1 * 100

% Change QMT Day 1 to Day 7 = (QMT Value Day 7 – QMT Value Day 1) / QMT Value Day 1 * 100

2.5.3 Correlation Ultrasound and Functional Capacity

To describe the relationship between muscle morphology and functional capacity, we investigated whether admission muscle mass could predict patients' functional independence at discharge. This was done by calculating the Spearman's rank correlation coefficient between the maximum CSA, RF, and QMT at ICU admission and Mobility-Transfer-Barthel at ICU discharge. Since patient inclusion required functional independence prior to admission, we assessed whether admission muscle mass influenced the degree of decline in the Mobility-Transfer-Barthel score. Therefore, correlation coefficients between the maximum ICU admission CSA, RF, and QMT and total Mobility-Transfer-Barthel decline from admission to discharge were calculated. Lastly, to account for differences in muscle mass on admission, a potential relationship between total absolute change of muscle mass and Mobility-Transfer-Barthel was explored. Relation was tested by correlating total absolute change on ultrasound with total decline on Mobility-Transfer-Barthel. For our primary outcome, only the subcategories transfer and mobility on level surfaces at hospital discharge were applied (maximum score of 30 points).

For a first subgroup analysis, patients were categorized by degree of functional decline: mild (10 points), moderate (15-20 points), or strong (25-30 points) decline on Mobility-Transfer-Barthel. The values of absolute change in CSA, RF, and QMT were then plotted with box plots comparing patients with mild, moderate, and strong decline in mobility. For a second subgroup analysis, patients were categorized by whether they had complete (0 points) or incomplete dependence (> 0 points) on transfer and mobility at the time of discharge. A Mann Whitney U Test was then applied to test for a significant difference in absolute change of ultrasound values (CSA, RF, and QMT) between completely and incompletely dependent patients.

2.5.4 Ultrasound and CT Imaging

Lastly, we compared both ultrasound CSA and thickness measurements with the gold standard, abdominal skeletal muscle CT imaging, in two steps. First, in all patients with a CT scan and an ultrasound upon ICU admission, values of total CT skeletal muscle area (SMA in cm^2) were explored for correlation with ultrasound values. Spearman's rank correlation coefficient was calculated for CSA, RF, and QMT, respectively. Secondly, because established CT cut-off values exist, the discriminative ability of ultrasound CSA, RF, and QMT measurements to detect low muscle mass was tested. For this, patients with a CT scan were stratified above and below gender-specific CT thresholds (men <52.4 cm^2/m^2 and women <38.5 cm^2/m^2). Differences in muscle mass measured by ultrasound were compared between CT groups for statistical significance using the Mann Whitney U Test.

3. Results

3.1 Patient Baseline Characteristics

Patient baseline characteristics are shown in Table 3.1. 42 (61.8%) of the 68 patients were male. Patient median age was 66 [IQR 55,77] years. The overall burden of prior comorbidities on the patient population was low, with a median CCI of 0 [IQR 0,2]. The main reason for ICU admission was non-traumatic brain injury in 19 (27.9%) patients, followed by respiratory failure in 14 (20.6%) patients. Upon ICU admission, the treating medical department to which most patients belonged was neurology (35%), followed by neurosurgery (31%).

Table 3.1 Patient characteristics

Patient characteristics	N= total patients (68)
Gender (n (%))	
male	42 (61.8)
female	26 (38.2)
Age (median [IQR])	66 [51, 77]
BMI (median [IQR])	24.8 [23.2, 27.0]
Admission from (n (%))	
Home	44 (64.7)
Other hospital	24 (35.3)
GCS (median [IQR])	9 [6, 14]
APACHE (median [IQR])	16 [10, 19]
SOFA (median [IQR])	7 [5, 8]
CCI (median [IQR])	0 [0, 2]
ICU Admission reason (n (%))	
Respiratory failure	14 (20.6)
Cardiovascular	2 (3.0)
Sepsis	6 (8.8)
Polytrauma without TBI	8 (11.8)
Traumatic brain injury (TBI)	12 (17.6)
Elective Surgery	2 (2.9)
Non-traumatic brain injury	19 (27.9)
other	14 (20.6)
Department (n (%))	
Neurosurgery	21 (30.9)
Ear Nose Throat	3 (4.4)
Abdominal Surgery	6 (8.8)
Vascular Surgery	4 (5.9)
Trauma Surgery	7 (10.3)
Internal Medicine	1 (1.5)
Neurology	24 (35.3)
Other	2 (2.9)

Descriptive statistics reported as median with interquartile ranges (IQR) and categorical variables as frequencies and percentages. *BMI* body mass index *GCS* Glasgow Coma Scale *APACHE* Acute Physiology and Chronic Health Evaluation *SOFA* Sequential Organ Failure Assessment *CCI* Charlson Comorbidity Index

3.2 Bland-Altman Plot Analysis

Bland-Altman plot analyses comparing agreement between measurements of CSA, RF, and QMT of the left and right leg in those patients with bilateral ultrasound assessment are presented in

figures 3.1, 3.2, and 3.3. For CSA, the mean difference between left and right measurement values lied slightly below zero (represented by the central dashed line in Figure 3.1).

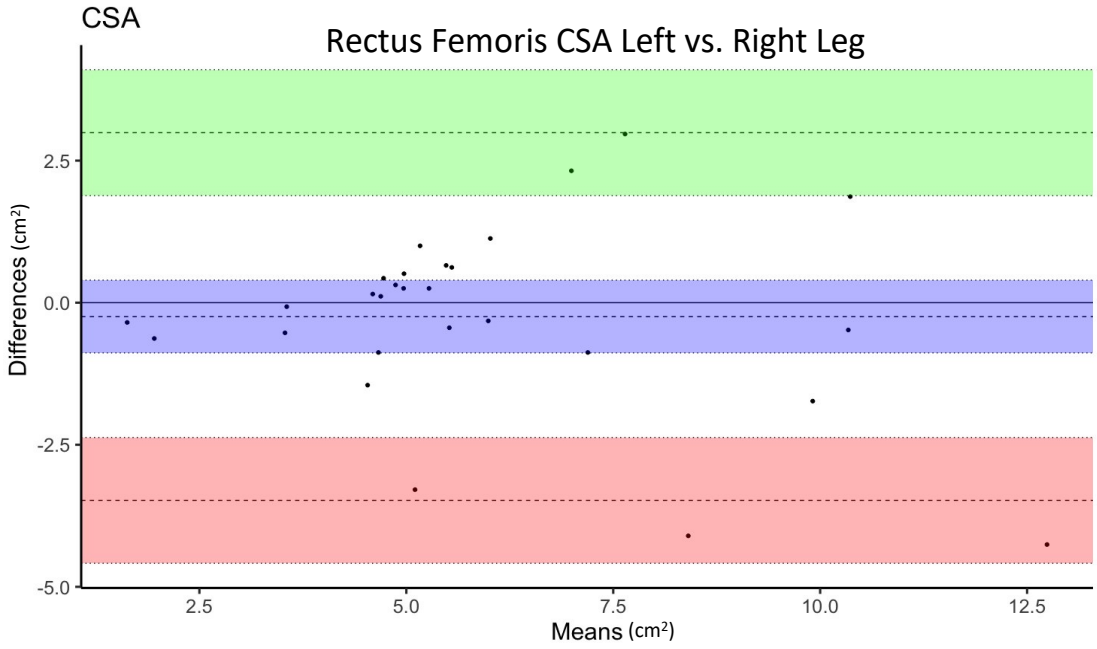

Figure 3.1 Bland-Altman plot analysis of rectus femoris cross-sectional area (CSA) values. The mean of the left and right measurement value for each patient with bilateral assessment was plotted against the absolute measurement difference between left and right values in cm^2. The green and red bars represent the confidence intervals two standard deviations above and below the mean difference between the left and right leg. Only patients with the measurement site of 3/5 distance from the anterior superior iliac spine (ASIS) to the upper border of the patella had bilateral ultrasound assessment, and were thus included in Bland-Altman analysis.

For measurements of the RF thickness, the mean difference between left and right measurement values lied at zero (see Figure 3.2).

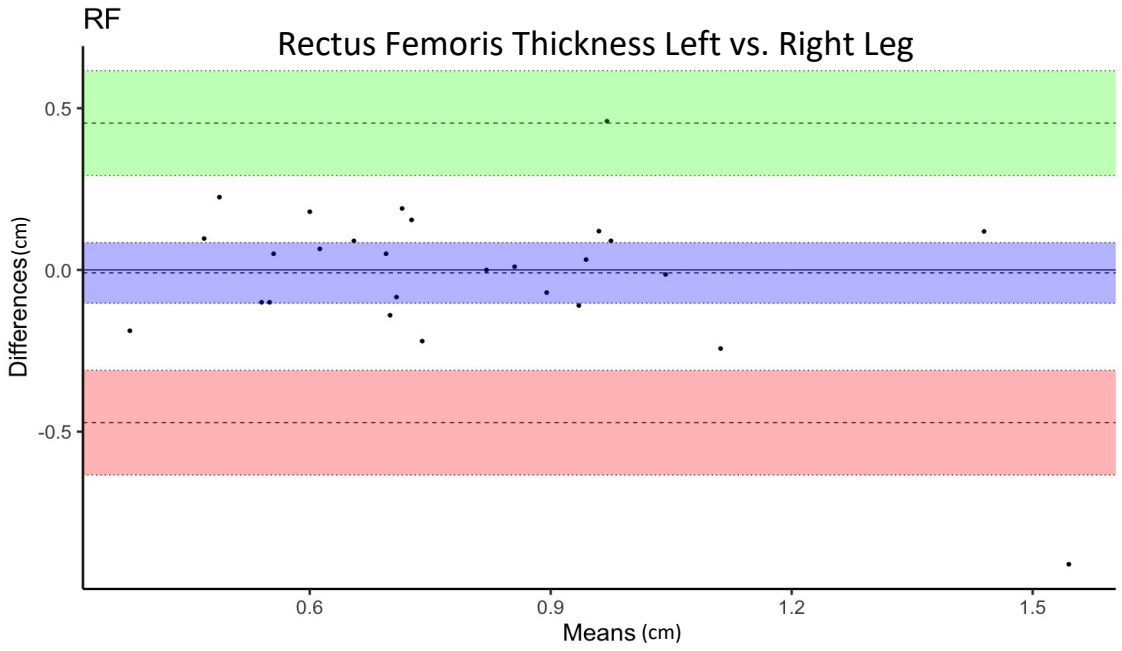

Figure 3.2 Bland-Altman plot analysis of rectus femoris thickness (RF) values. The mean of the left and right measurement value for each patient with bilateral assessment was plotted against the absolute measurement difference between left and right values in cm. The green and red bars represent the confidence intervals two standard deviations above and below the mean difference between the left and right leg. Only patients with the measurement site of 3/5 distance from the anterior superior iliac spine (*ASIS*) to the upper border of the patella had bilateral ultrasound assessment, and were thus included in Bland-Altman analysis.

For QMT, the mean difference between left and right measurement values lied at zero (see Figure 3.3). No proportional bias was observed across the three measurements of CSA, RF, and QMT. CSA and QMT measurement values showed greater variance between the left and right leg compared to the RF measurement. Larger mean measurement values did not influence the degree of variance between the left and right leg measurement values.

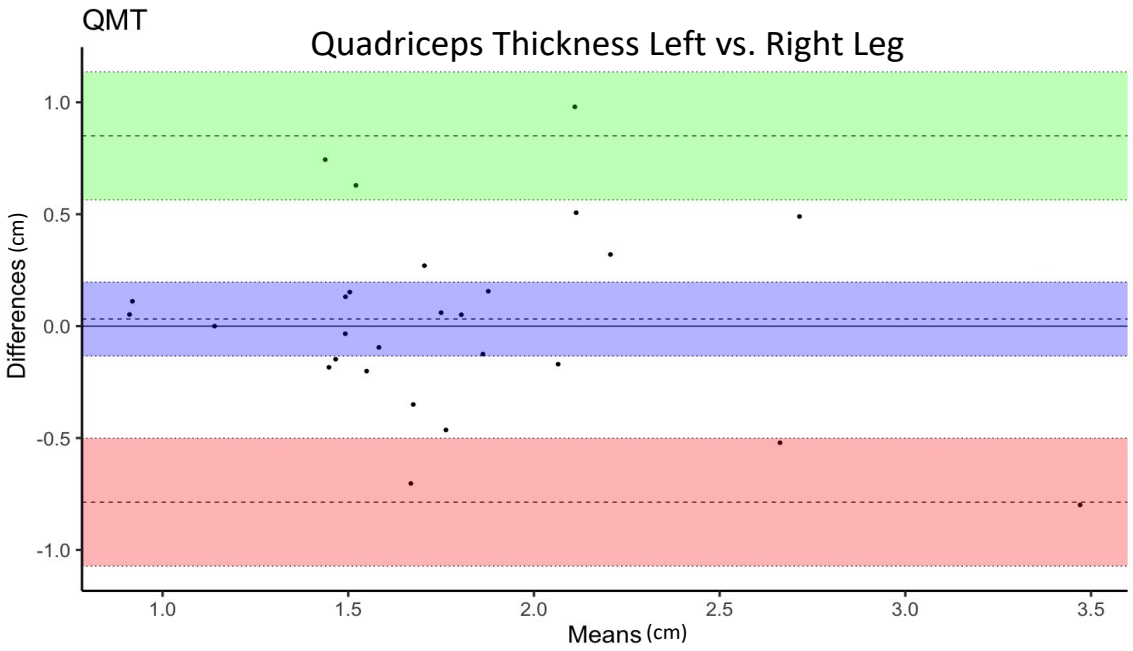

Figure 3.3 Bland-Altman plot analysis of quadriceps muscle thickness (QMT) values. The mean of the left and right measurement value for each patient with bilateral assessment was plotted against the absolute measurement difference between left and right values in cm. The green and red bars represent the confidence intervals two standard deviations above and below the mean difference between the left and right leg. Only patients with the measurement site of 3/5 distance from the anterior superior iliac spine (ASIS) to the upper border of the patella had bilateral ultrasound assessment, and were thus included in Bland-Altman analysis.

3.3 Change in CSA, RF, and QMT from Admission to Discharge

The median absolute change in ultrasound measurements from ICU admission to ICU discharge using the maximum, mean, and minimum values for CSA, RF, and QMT are shown in Table 3.2. There was an overall decline in the median absolute values of all three measurements from admission to discharge. Decline was irrespective of whether the maximum, mean, or minimum values to each measurement time point were used for calculation. Median absolute CSA decline using the mean CSA value was -1.69 cm^2 [IQR -2.53, -0.53]. Median QMT and RF thickness declined by 0.53 cm [IQR -0.74, -0.26] and 0.27 cm [IQR -0.36, -0.01], respectively when using mean values for calculation. Differences in calculated absolute CSA change when using maximum and minimum values was 0.2 cm^2. Patients that did not have an ultrasound assessment on the day of ICU discharge or that had inadequate image quality are counted as missing in Table 3.2.

Table 3.2 Absolute change in ultrasound measurements presented as median [IQR]				
			Overall	Missing (n)
n			68	
CSA (cm^2)	Max	ICU admission	5.26 [3.72, 6.96]	9
		ICU discharge	3.82 [3.16, 5.06]	32
		Absolute change	-1.74 [-2.93, -0.39]	37
	Mean	ICU admission	5.11 [3.60, 6.91]	9
		ICU discharge	3.82 [2.90, 4.97]	32
		Absolute change	-1.69 [-2.53, -0.53]	37
	Min	ICU admission	4.72 [3.54, 6.36]	9
		ICU discharge	3.75 [2.84, 4.68]	32
		Absolute change	-1.54 [-2.21, -0.51]	37
QMT (cm)	Max	ICU admission	1.80 [1.45, 2.14]	9
		ICU discharge	1.30 [1.06, 1.62]	29
		Absolute change	-0.53 [-0.76, -0.25]	33
	Mean	ICU admission	1.71 [1.43, 2.09]	9
		ICU discharge	1.30 [1.05, 1.58]	29
		Absolute change	-0.53 [-0.74, -0.26]	33
	Min	ICU admission	1.62 [1.32, 1.92]	9
		ICU discharge	1.30 [1.02, 1.48]	29
		Absolute change	-0.53 [-0.76, -0.26]	33
RF (cm)	Max	ICU admission	0.80 [0.60, 0.96]	10
		ICU discharge	0.57 [0.44, 0.77]	29
		Absolute change	-0.23 [-0.41, 0.00]	33
	Mean	ICU admission	0.76 [0.56, 0.94]	10
		ICU discharge	0.57 [0.44, 0.75]	29
		Absolute change	-0.27 [-0.36, -0.01]	33
	Min	ICU admission	0.74 [0.53, 0.93]	10
		ICU discharge	0.53 [0.44, 0.69]	29
		Absolute change	-0.27 [-0.35, -0.07]	33

Absolute change of measurement values from ICU admission to discharge presented as median with interquartile ranges [IQR]. If left and right leg value available, max, min, or mean value used. If only one leg value available, same value used for max, min, and mean calculation of absolute change. Patients without ultrasound assessment on date of ICU discharge reported as missing. Note: 19/28 patients in the NICU cohort without ultrasound assessment on date of discharge included under missing. *CSA* Rectus femoris cross-sectional area (cm^2) *RF* Rectus femoris thickness (cm) *QMT* Quadriceps muscle thickness (cm) *max* largest measurement value at admission or discharge *mean* average of left and right leg measurement value *min* smallest measurement value at admission or discharge

The calculated median percentage change from admission to discharge across all measured images for CSA, RF, and QMT is shown in Table 3.3. Overall, the percentage decline from admission continues to increase in magnitude from Day 7 to ICU discharge. CSA showed the greatest percentage decline after seven days taken across all right leg measurements [-14.14%]. Values of median percentage change showed greater variation between left and right at Day 7 than at ICU discharge.

Table 3.3 Percentage change of ultrasound measurements presented as median [IQR]

	Number of measurement values	% Change to Day 7 (median [IQR])	% Change to last US (median [IQR])	% Change to discharge US (median [IQR])
CSA left leg	34*	-8.39 [-20.60, 2.11]	-24.04 [-38.86, -8.25]	-25.76 [-35.47, -10.04]
QMT left leg	32*	-8.09 [-25.57, 6.52]	-14.24 [-26.45, 3.76]	-21.44 [-30.50, -1.74]
RF left leg	32*	-8.82 [-25.54, -11.70]	-22.22 [-32.38, 15.96]	-28.36 [-44.16, -11.25]
CSA right leg	62	-14.14 [-31.18, 7.20]	-21.19 [-41.27, -0.71]	-25.10 [-45.49, 4.57]
QMT right leg	61	-10.11 [-24.75, 0.00]	-21.91 [-40.49, -8.71]	-27.78 [-42.77, -12.33]
RF right leg	61	-12.82 [-23.46, 9.61]	-20.18 [-38.80, -2.00]	-27.38 [-43.96, 0.00]

Median percentage change with interquartile range [IQR] from admission to Day 7, to discharge, or to last ultrasound assessment calculated for CSA, RF, and QMT across all measured images, regardless of image quality. If ultrasound was not performed on the day of ICU discharge, percentage change was calculated to the last available assessment in the ICU. *Due to differing ultrasound protocols between studies, bilateral ultrasound was not performed in all patients. Number of measurement values represents the number of paired ultrasound values for calculation of percentage change. *US* Ultrasound *IQR* Interquartile range *CSA* Rectus femoris cross-sectional area (cm^2) *RF* Rectus femoris thickness (cm) *QMT* Quadriceps muscle thickness (cm)

3.4 Decline of Muscle Mass on Ultrasound and Functional Capacity

The absolute change in functional capacity in survivors from ICU admission to hospital discharge is presented in Figure 3.4. Although there was an overall decline in median ultrasound measurement values from ICU admission to discharge, not all patients followed this pattern. Patients with an increase in measurement values were seen for all three measurements (CSA, RF, and QMT). The vast majority of patients had a maximum Mobility-Transfer-Barthel score (30 points) at admission. Few patients retained maximum mobility by the time of hospital discharge, with the majority showing a decline over ICU and hospital stay.

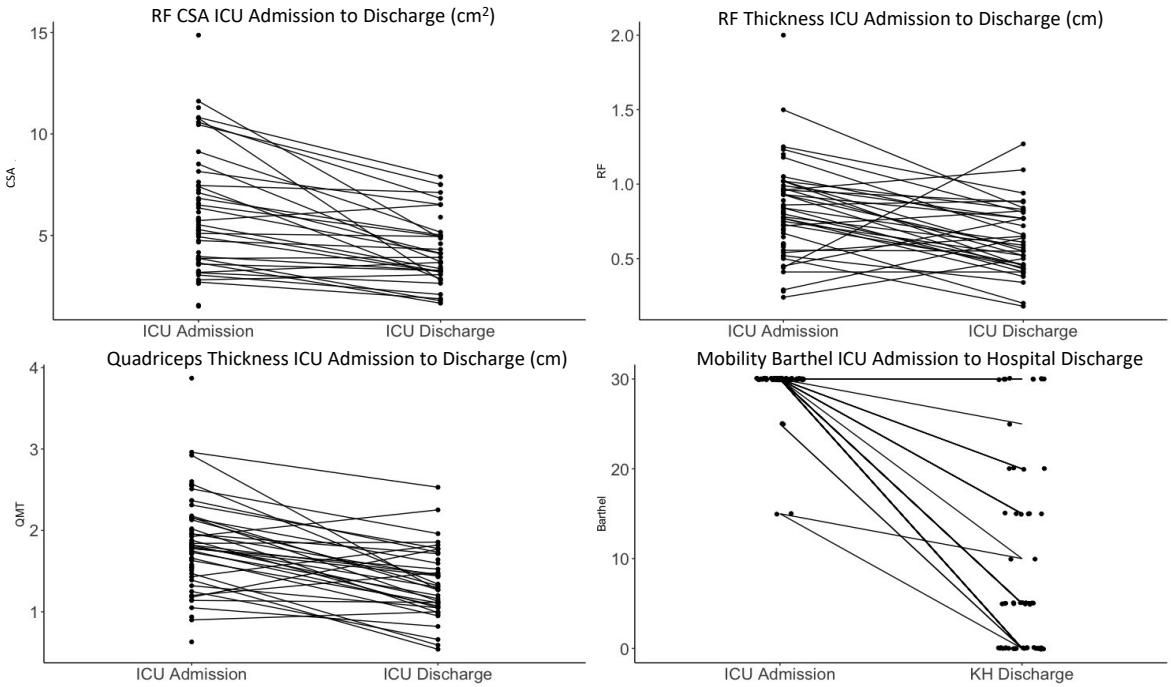

Figure 3.1 Plotted change in ultrasound measurement values and Mobility-Transfer-Barthel from ICU admission to discharge. Upper left: change in rectus femoris cross-sectional area (CSA) in cm^2. Upper right: change in rectus femoris thickness (RF) in cm. Lower left: change in quadriceps muscle thickness (QMT) in cm. Lower right: change in Mobility-Transfer-Barthel from ICU admission to hospital discharge. A maximum score of 30 points represents complete functional independence on transfer (bed to chair and back) and mobility on level surfaces (walking with or without gait-aid).

Table 3.4 and 3.5 list Spearman's rank correlation coefficients (ρ) between ultrasound measurement values and the Mobility-Transfer-Barthel. There was no correlation between maximum CSA, RF, and QMT on admission and Mobility-Transfer-Barthel score at hospital discharge (ρ=-0.01, p=0.918; ρ=0.032, p=0.814; ρ=0.064, p=0.629, respectively). Additionally, there was no correlation between the change in CSA, RF, and QMT from hospital admission to discharge and total change in Mobility-Transfer-Barthel score (ρ=-0.03, p=0.883; ρ=-0.12, p=0.510; ρ=0.08, p =0.630, respectively).

Table 3.4 Correlation between ultrasound measurement values and Mobility-Transfer-Barthel

	Mobility-Transfer-Barthel (hospital discharge)	
	Spearman's correlation (ρ)	p value
Max CSA Admission (cm^2)	-0.01	0.918
Max RF Admission (cm)	-0.032	0.814
Max QMT Admission (cm)	0.064	0.629

CSA Rectus femoris cross-sectional area (cm^2) *RF* Rectus femoris thickness (cm) *QMT* Quadriceps muscle thickness (cm) *max* largest measurement value at admission *Mobility-Transfer-Barthel* Independence on transfer (bed to chair and back) and mobility on level surfaces (walking with or without gait-aid) at hospital discharge

Table 3.5 Correlation between changes in ultrasound values and Mobility-Transfer-Barthel

	Change in Mobility-Transfer-Barthel (admission to hospital discharge)	
	Spearman's correlation (ρ)	p value
Change in max CSA (cm^2)	-0.03	0.883
Change in max RF (cm)	-0.12	0.510
Change in max QMT (cm)	0.08	0.630

CSA Rectus femoris cross-sectional area (cm^2) *RF* Rectus femoris thickness (cm) *QMT* Quadriceps muscle thickness (cm) *max* largest measurement value at admission *Mobility-Transfer-Barthel* Absolute change in independence on transfer (bed to chair and back) and mobility on level surfaces (walking with or without gait-aid) from admission to hospital discharge

3.5 Subgroup Analysis

In a first subgroup analysis, values of absolute change (using the maximum for CSA, RF, and QMT) were plotted with boxplots for strong, moderate, and mild functional decline. There was no significant difference in the median change of measurement values for CSA, RF, and QMT between groups (see Figures 3.5, 3.6, and 3.7). All three measurements declined, irrespective of the degree of functional decline.

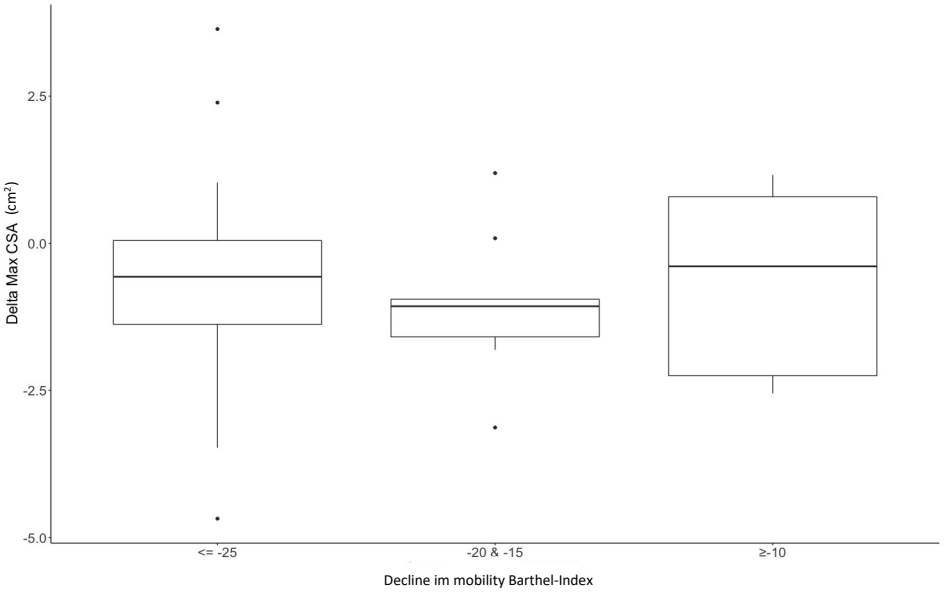

Figure 3.2 Change (delta) in rectus femoris cross-sectional area (CSA in cm^2) from admission to ICU discharge categorized by degree of decline in functional capacity on Mobility-Transfer-Barthel. Severe decline: $\leq$ -25 points; moderate decline: -20 to -15 points; mild decline: $\geq$ -10 points. Boxes outline the interquartile ranges (IQR), in which 50% of measurement values lie. Single plotted values lie outside of 1.5 times the IQR. The boldened line within each box represents the median absolute change on CSA ultrasound.

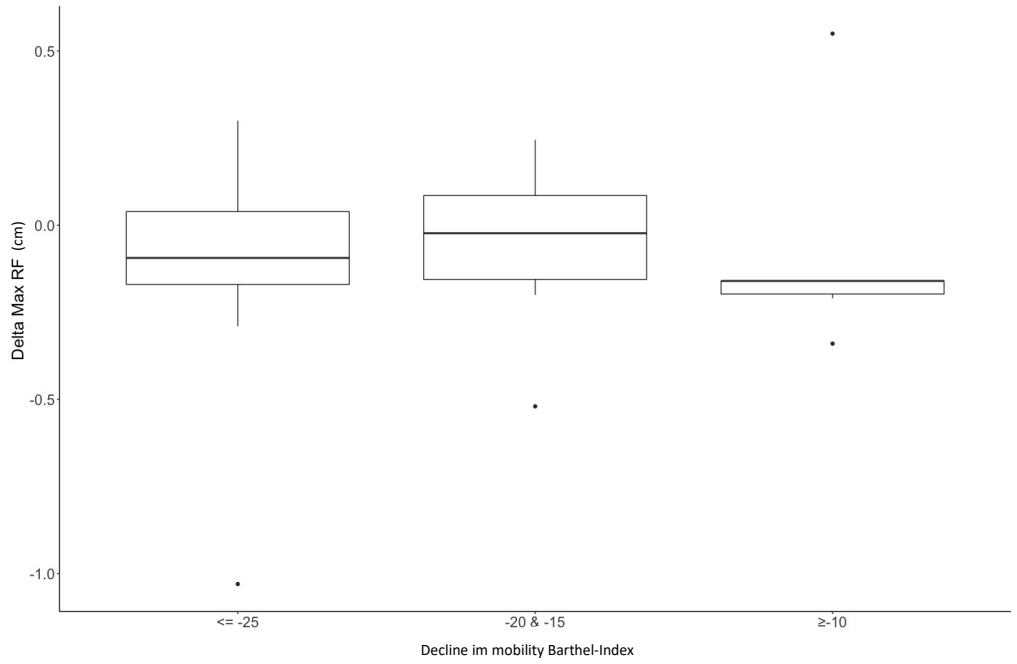

Figure 3.3 Change (delta) in rectus femoris thickness (RF in cm) categorized by degree of decline in functional capacity on Mobility-Transfer-Barthel. Severe decline: ≤ -25 points; moderate decline: -20 to -15 points; mild decline: ≥ -10 points. Boxes outline the interquartile range (IQR), in which 50% of measurement values lie. Single plotted values lie outside of 1.5 times the IQR. The boldened line within each box represents the median absolute change on RF thickness ultrasound.

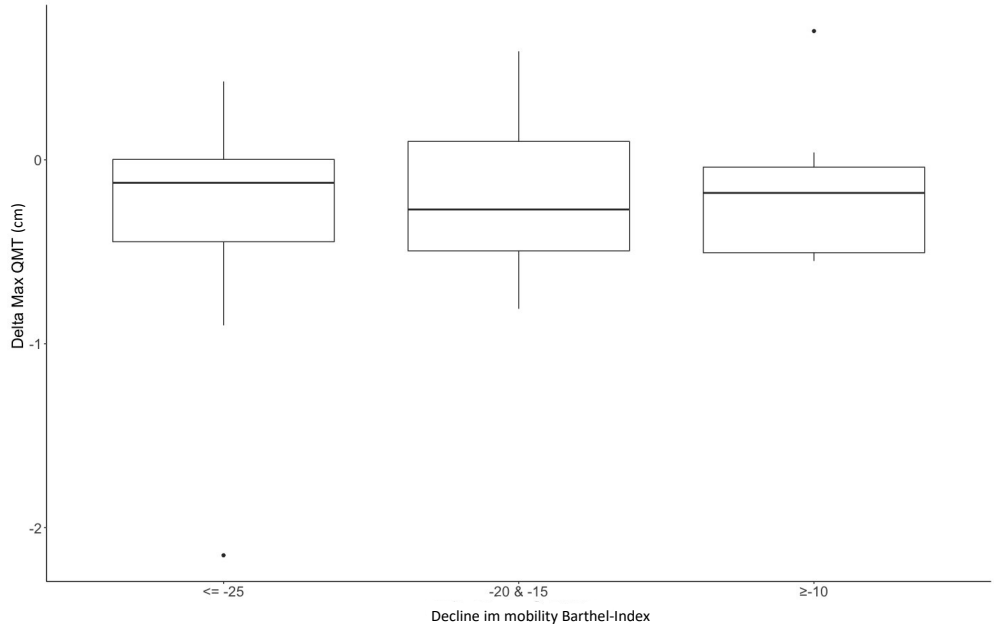

Figure 3.4 Change (delta) in quadriceps muscle thickness (QMT in cm) categorized by degree of decline in functional capacity on Mobility-Transfer-Barthel. Severe decline: ≤ -25 points; moderate decline: -20 to -15 points; mild decline: ≥ -10 points. Boxes outline the interquartile range (IQR), in which 50% of measurement values lie. Single plotted values lie outside of 1.5 times the IQR. The boldened line within each box represents the median absolute change on QMT ultrasound.

In a second subgroup analysis, patients were further categorized by whether they were completely (-30 points) or incompletely (> -30 points) dependent at discharge, defined by Mobility-Transfer-Barthel. 23 patients had zero points on the Mobility-Transfer-Barthel (see Table 3.6). There was no statistically significant difference in the absolute change of muscle mass measurements between the groups with and without complete functional dependence. This held true regardless of whether maximum, minimum, or mean values were used for calculation of absolute change from baseline to ICU discharge (p values listed in Table 3.6).

Table 3.6 Change on ultrasound by complete vs. incomplete dependency

	-30	>-30	p
n	23	44	
Delta max CSA to discharge in cm^2 (median [IQR])	-0.52 [-2.17, 0.04]	-1.90 [-2.94, -0.73]	0.296
Delta max RF to discharge in cm (median [IQR])	-0.22 [-0.32, -0.02]	-0.31 [-0.42, 0.00]	0.596
Delta max QMT to discharge in cm (median [IQR])	-0.44 [-0.55, -0.08]	-0.55 [-0.85, -0.27]	0.228
Delta min CSA to discharge in cm^2 (median [IQR])	-0.52 [-2.17, 0.12]	-1.74 [-2.21, -0.73]	0.317
Delta min RF to discharge in cm (median [IQR])	-0.26 [-0.33, -0.14]	-0.28 [-0.36, 0.00]	0.971
Delta min QMT to discharge in cm (median [IQR])	-0.37 [-0.56, -0.17]	-0.53 [-0.85, -0.31]	0.265
Delta mean CSA to discharge in cm^2 (median [IQR])	-0.54 [-2.17, 0.08]	-1.81 [-2.68, -0.76]	0.317
Delta mean RF to discharge in cm (median [IQR])	-0.25 [-0.31, -0.06]	-0.31 [-0.37, 0.00]	0.675
Delta mean QMT to discharge in cm (median [IQR])	-0.42 [-0.54, -0.13]	-0.55 [-0.81, -0.32]	0.214

Comparison of patients with complete (-30 points) and incomplete dependency (> - 30 points) in subcategories transfer and mobility at hospital discharge with the Mann Whitney U Test. Completely dependent patients were unable to perform transfer from bed to chair (including with assistance) and were immobile on level surfaces. All values for absolute change (delta) on ultrasound from admission to discharge presented as median with interquartile ranges [IQR]. If a left and right leg value was available, max, min, or mean value used. If only one leg value was available, the same value used for max, min, and mean calculation of absolute change. *IQR* Interquartile range *CSA* Rectus femoris cross-sectional area (cm^2) *RF* Rectus femoris thickness (cm) *QMT* Quadriceps muscle thickness (cm)

3.6 Comparison of Ultrasound with CT Measurements

Lastly, we compared the muscle imaging modalities of ultrasound and CT in our cohort of 68 patients with a minimum stay of 7 days. After quality control of all CT images (see page 25 for criteria), 22 patients had both a CT scan and an ultrasound assessment for comparison at ICU admission. Results of the comparisons between techniques are displayed in Figure 3.8. The correlation between skeletal muscle area on CT and CSA on ultrasound (ρ=0.619 p=0.003) was stronger than with RF (ρ=0.332;p=0.12) and QMT (ρ=0.453;p=0.03).

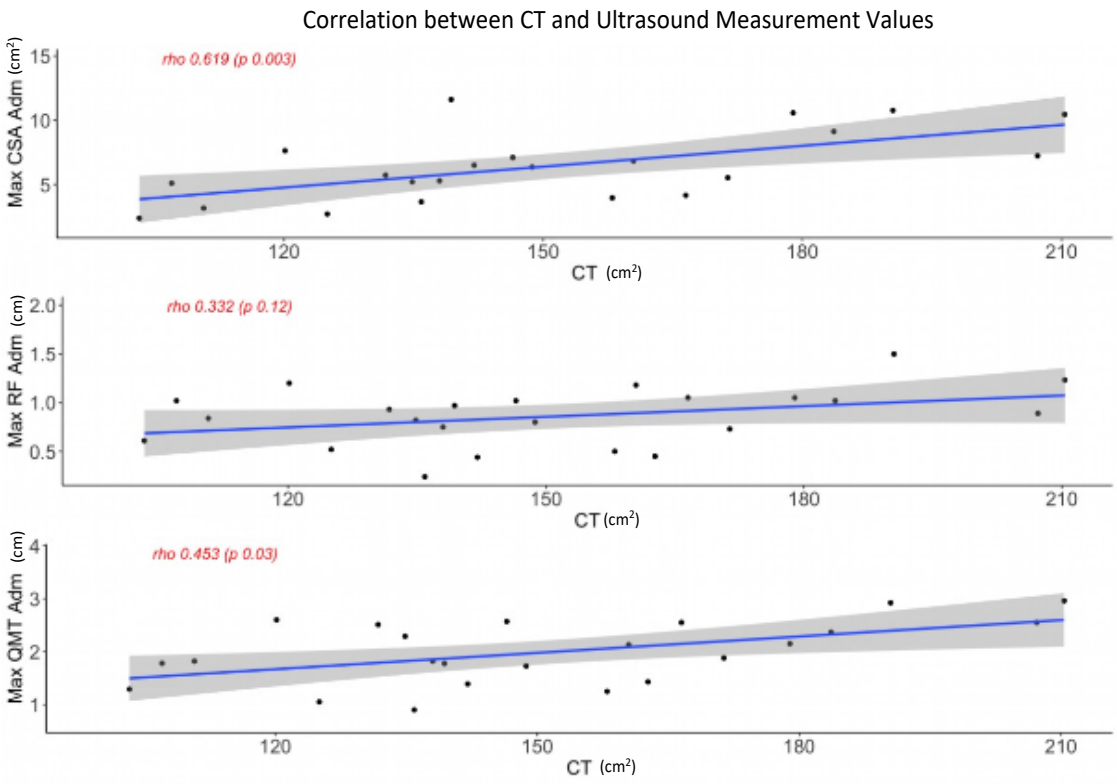

Figure 3.5 Correlation between CT total skeletal muscle area (cm^2) and ultrasound measurement values (CSA,RF, QMT) at ICU admission. Comparison of ultrasound with CT was possible in 22 of 68 patients. Upper panel: maximum admission rectus femoris cross-sectional area (CSA in cm^2); mid panel: maximum admission rectus femoris thickness (RF in cm); lower panel: maximum admission quadriceps muscle thickness (QMT in cm). In red: Spearman's rank correlation coefficient (rho) with p values.

In order to compare results between both imaging modalities, we tested the discriminative ability of ultrasound to detect sarcopenic patients below the CT cut-off (skeletal muscle index: men <52.4 cm^2/m^2 and women <38.5 cm^2/m^2). Results are presented in Table 3.7. Of 27 patients with a CT during hospital stay, 9 patients were below (labeled "below" in Table 3.7) and 18 patients were above (labeled "above" in Table 3.7) the sarcopenia cut-off on their first scan. Ultrasound values tended to be smaller for CSA, RF, and QMT in patients below the CT cut-off. Although CSA on ultrasound showed a stronger correlation with CT SMA on admission than RF and QMT, there was no significant difference in ultrasound measurement values between patients above and below the CT cut-off. This held true, irrespective of whether the maximum, mean, or minimum measurement values were used for calculation.

Table 3.7 Ultrasound measurement values stratified by sarcopenia on CT

Sarcopenia Cut-off	Below	Above	p
n	9	18	
Max CSA admission (mean (SD))	5.88 (1.26)	6.61 (3.11)	0.586
Max RF admission (mean (SD))	0.76 (0.30)	0.90 (0.30)	0.322
Max QMT admission (mean (SD))	1.81 (0.54)	2.06 (0.61)	0.361
Min CSA admission (mean (SD))	5.59 (1.21)	6.03 (2.63)	0.697
Min RF admission (mean (SD))	0.68 (0.22)	0.87 (0.27)	0.123
Min QMT admission (mean (SD))	1.66 (0.41)	1.96 (0.53)	0.205
Mean CSA admission (mean (SD))	5.73 (1.22)	6.32 (2.81)	0.629
Mean RF admission (mean (SD))	0.72 (0.25)	0.89 (0.28)	0.206
Mean QMT admission (mean (SD))	1.74 (0.45)	2.01 (0.56)	0.270
ICU Mobility-Transfer Barthel (median [IQR])	10.00 [5.00, 20.00]	5.00 [0.00, 8.75]	0.059
ICU Mobility-Transfer-Barthel (%)			0.177
0	1 (11.1)	8 (44.4)	
5	3 (33.3)	5 (27.8)	
10	1 (11.1)	1 (5.6)	
15	1 (11.1)	3 (16.7)	
20	2 (22.2)	0 (0.0)	
25	0 (0.0)	1 (5.6)	
30	1 (11.1)	0 (0.0)	

Ultrasound measurements were tested for discriminative ability to detect sarcopenic patients below the CT cut-off skeletal muscle index (SMI) using the Mann Whitney U Test. SMI cutoff: men <52.4 cm^2/m^2 and women <38.5 cm^2/m^2. Maximum, mean, and minimum ultrasound admission values used for analysis are presented as mean with standard deviation (SD). Comparison of patients at different scores for individual subcategories of the Mobility-Transfer-Barthel (transfer and mobility on level surfaces) were stratified by low muscle mass. If ultrasound was only performed on one leg, the same value was used for calculation towards maximum, mean, and minimum. *IQR* Interquartile range *CSA* Rectus femoris cross-sectional area (cm^2) *RF* Rectus femoris thickness (cm) *QMT* Quadriceps muscle

4. Discussion

4.1 Principal Findings

We aimed to test the predictive capability of different commonly used ultrasound measurements for functional capacity at discharge in our cohort of critically ill patients. We found a strong overall decline in the level of patients' functional capacity, with 23 of the 68 patients experiencing a complete loss of mobility. Although we observed overall decline in functional independence and muscle mass on ultrasound, we found no correlation between muscle morphology and functional outcome in our cohort. Thus, muscular status at admission was not a predictor of functional independence at discharge. Further, after categorizing patients by the degree of loss in functional independence, we found no significant difference in measured muscle mass between groups. Lastly, we compared ultrasound with CT imaging in patients with a performed abdominal scan, finding that skeletal muscle area on CT correlated more with ultrasound cross-sectional area than with thickness measurements. In this small cohort of patients with an abdominal CT, ultrasound showed poor discriminative ability to detect low muscle mass in patients defined as sarcopenic on CT.

4.2 Comparison with other Studies

4.2.1 Ultrasound

The loss of functional independence in activities of daily living patients experience after critical illness is multifactorial. Factors such as older age and longer ICU length of stay have been associated with higher disability 7 days post-discharge (Herridge, Chu et al. 2016). This has subsequently been associated with the trajectory of recovery and 1-year mortality (Herridge, Chu et al. 2016). Yet, understanding of muscular atrophy as a driving force of reduced strength and subsequent functional disability remains incomplete. Specifically, the role of ultrasound as a tool to predict disability remains unclear due to conflicting evidence throughout the literature (Mayer, Thompson Bastin et al. 2020).

Table 4.1 Comparison of results with similar studies

	Study population	Ultrasound parameter	Admission values	7-day percent decline	Relation to functional outcome
Results of current study	68 previously functionally independent critically ill patients	CSA, RF, QMT at ICU admission, Day 7, weekly until ICU discharge	Admission Median CSA: 5.11 cm^2 Median QMT*: 1.71 cm Median RF*: 0.76 cm	Median % decline after 7 days** CSA: -14.1% QMT: -10.1% RF: -12.8%	-No correlation admission CSA, RF, or QMT with mobility subcategories of Barthel-Index at hospital discharge -No correlation CSA, RF, or QMT absolute change with decline on mobility subcategories from admission to hospital discharge
Palakshappa et al.	29 critically ill patients with sepsis complicated by shock or respiratory failure	CSA and QMT at ICU admission and Day 7 measured at 2/3 distance from the ASIS to the upper patellar border	Admission Median CSA: 4.33 cm^2 Median QMT: 2.23 cm	Median % decline after 7 days CSA: - 23.2% QMT: - 17.9%	-Moderate, not statistically significant correlation between daily % reduction in CSA and PFIT score ($\rho = 0.4$, $p = 0.10$) - No correlation between admission or Day 7 CSA with PFIT score on Day 7 - Baseline, Day 7, and daily % reduction in QMT showed no correlation with PFIT score
Puthucheary et al.	19 critically ill patients with mechanical ventilation for a minimum of two days	CSA and QMT on Day 1, 7, and 10 measured at 3/5 distance from the ASIS to the upper patellar border	Not included	% decline after 7 days CSA: - 13.0% QMT: -5.9%	MRC strength test on Day 10 - Decline in CSA greater in those with knee extensor weakness at Day 10 than without (20.7% vs. 8.4%; $p = 0.012$) - Decline in QMT did not differ between group with and without knee extensor weakness at Day 10 (12.6% vs. 12.1%; $p = 0.95$)
Parry et al.	22 critically ill patients with mechanical ventilation for a minimum of two days	CSA and RF thickness, VI thickness on ICU admission, Day 3, 5, 7, 10, at awakening, and ICU discharge measured at 2/3 distance from the ASIS to the upper patellar border	Admission Mean CSA: 4.42 cm^2 Mean RF: 2.44 cm Mean VI: 1.91 cm	% decline after 7 days CSA: - 16.8% RF: - 24.9% VI: - 20.0%	MRC, PFIT, IMS score at awakening (performed median 9 days after admission) and ICU discharge - CSA moderately correlated with PFIT ($r = 0.71$ $p = 0.02$) and IMS ($r = 0.68$ $p = 0.03$) at ICU discharge - Strong correlation VI thickness with PFIT ($r = 0.82$ $p < 0.001$) and IMS ($r = 0.84$ $p < 0.001$) at ICU discharge - RF correlated ($r = 0.58$ $p = 0.03$) with PFIT and IMS ($r = 0.63$ $p = 0.02$) at ICU discharge
Mayer et al.	41 critically ill patients with acute respiratory failure or sepsis	CSA, RF, and QMT on Day 1, 3, 5, 7 measured at 2/3 distance from the ASIS to the upper patellar border	Admission Mean CSA: 2.99 cm^2 Mean RF: 0.98 cm Mean QMT: 2.04 cm	% decline after 7 days CSA: -18.5% RF: - 20.1% QMT: -14.5%	-Admission CSA, RF and 7-day % decline in CSA, RF showed weak correlation with 5 x sit-to-stand test, 4-meter gait speed, and 6-minute walk distance scores at hospital discharge

* ultrasound performed using maximum compression ** percentage decline of right leg measurement values for purpose of comparison

PFIT Physical Function in the ICU tests assistance (sit to stand), cadence (steps/min), shoulder (flexion) and knee (extension) strength

MRC sum score tests the strength of different upper and lower extremity muscle groups bilaterally *r* Pearson's correlation coefficient

IMS Intensive care unit mobilization scale assesses the highest level of mobilization reached *VI* Vastus intermedius muscle

ASIS anterior superior iliac spine *CSA* cross-sectional area (cm^2) *RF* rectus femoris thickness (cm) *QMT* Quadriceps muscle thickness (cm)

Acknowledging the differences in measurement protocols, studies vary in the reported 7-day decline of muscle cross-sectional area and thickness (see Table 4.1). Differences in baseline functional status, comorbidities, severity of illness, as well as admission muscle mass may influence the degree of muscle wasting in the ICU. For example, Palakshappa et al. only included patients with sepsis complicated by respiratory failure or shock in their study. Their severely ill cohort was at high risk for muscle wasting and showed greater 7-day muscle decline than our cohort. Those studies with comparable cohorts to ours showed similar degree of 7-day muscle decline. It is clear though that multiple studies have confirmed muscle wasting to begin early in the course of critical illness (Puthucheary, Rawal et al. 2013, Parry, El-Ansary et al. 2015, Puthucheary, McNelly et al. 2017, Palakshappa, Reilly et al. 2018, Mayer, Thompson Bastin et al. 2020).

Some studies have shown one measurement to be more representative of muscle weakness than another (Puthucheary, McNelly et al. 2017). These results are challenging to compare with our study, as it is unclear whether low muscle strength consistently translates to low functional status. Others have found weak or no correlation to function for both cross-sectional area and thickness, similar to our data (see Table 4.1) (Mayer, Thompson Bastin et al. 2020). Additionally, assessments beyond one- or two-dimensional ultrasound measurements have shown more promising results: Mayer et al. found that muscle "power", the velocity with which patients performed a leg press against resistance, was a stronger independent predictor of performance on functional tests. As in our cohort, measured muscle mass on admission and the decline in rectus femoris thickness, quadriceps muscle thickness, and cross-sectional area to discharge were poor predictors of functional performance. In light of these results, the authors suggest that muscle power, which is required to overcome gravity when standing up from a chair, instead of muscle mass, could be a better predictor of functional outcome (Mayer, Thompson Bastin et al. 2020). While dynamic muscle power testing may be an objective method to assess muscular dysfunction, the required equipment may not be readily available. Further head-to-head comparisons between ultrasound and dynamic tests for predictive capability of functional performance at discharge are needed.

Lastly, our assessment of mobility by components of the Barthel-Index may not have fully captured muscular dysfunction as a result of muscle wasting. Also, the level of functional

independence our patients reached at discharge could have been influenced by the intensity of provided physical therapy after transfer from the ICU to the general ward. As early mobilization is critical to support functional independence, the level our patients reached may have been influenced by barriers such as pain, sedation, balance issues, or fear of falling. Although we did not factor these potential barriers into our analysis, other studies have reported this to be a common limitation when performing strength or functional tests.

4.2.2 Computer Tomography

Few studies have compared muscle ultrasound measurements with CT skeletal muscle area in critically ill patients. To the best of our knowledge, no study to date has compared both ultrasound cross-sectional area and thickness measurements with CT skeletal muscle area in a critically ill population, making ours the first. It remains unclear which ultrasound measurement is the best surrogate of whole-body skeletal muscle mass when CT skeletal muscle area is the reference. Whether cross-sectional area or thickness (with or without compression) of one muscle or multiple (upper and lower extremity) muscle groups correlates best with CT values in the critically ill is also unknown. Also, potentially different rates of muscle wasting for different muscle groups needs to be considered. Paris et al. assessed CT scans performed within 72 hours of maximally compressed ultrasound quadriceps thickness measurements in critically ill patients. As in our study, there was only a moderate correlation between quadriceps thickness and CT skeletal muscle area. The authors concluded that one thickness measurement may not be enough to accurately classify patients as having low whole-body muscle mass (Paris, Mourtzakis et al. 2017). This is concordant with our results, as quadriceps thickness measurements could not discriminate sarcopenic from non-sarcopenic patients in our cohort. This may be due to stronger muscle wasting in muscle groups captured on abdominal CT than on quadriceps ultrasound. Also, the ultrasound measurement protocol (with or without maximal compression) may affect comparison with CT. Another study performed by Fetterplace et al. retrospectively investigated whether maximally or minimally compressed ultrasound quadriceps thickness measurements could predict skeletal muscle area on an abdominal CT. Maximally compressed thickness measurements, as in our cohort, were smaller for patients defined as sarcopenic on CT. Interestingly, this did not hold true for thickness measurements performed without compression in their study. This finding supports

using maximal over minimal compression when performing ultrasound thickness measurements, as fluid may confound comparisons with CT. Further, ultrasound thickness measurements were predictive of skeletal muscle area on CT in their study. As was the issue in our cohort, repeat CT scans are often lacking. Thus, it remains unclear if changes in thickness on ultrasound are proportional to changes in skeletal muscle area on CT (Fetterplace, Corlette et al. 2021). This underscores how difficult it is to compare non-invasive measurements with CT measurements in critically ill patients. Lastly, the total number of muscle groups assessed with ultrasound may influence the comparison with CT imaging. Lambell et al. compared five different landmarks for ultrasound thickness measurements with CT skeletal muscle area. The authors found both upper-arm and quadriceps thickness measurements to strongly correlate with CT skeletal muscle area, concluding it may be important to assess multiple muscle groups for prediction of whole-body muscle mass. After adding the covariables age, sex, and Charlson Comorbidity Index to ultrasound thickness measurements, the authors substantially improved their prediction model for CT skeletal muscle area. Since we did not adjust for these covariables, our correlation between muscle thickness and CT skeletal muscle area was only moderate. Therefore, measurements of multiple muscle groups and adjustment for covariables may strengthen the correlation with CT skeletal muscle area (Lambell, Tierney et al. 2021). On the other hand, measuring multiple muscle groups may impact the practicality of bedside ultrasound in the ICU.

Rectus femoris cross-sectional area on ultrasound showed a stronger correlation with CT skeletal muscle area than muscle thickness in our cohort. This suggests ultrasound cross-sectional area could potentially be a better surrogate of whole-body muscle mass compared to thickness measurements. Further studies with larger sample sizes comparing both ultrasound measurements with CT are required. Should ultrasound cross-sectional area consistently show stronger correlation with CT skeletal muscle area, then it may become the standard measurement when performing muscle mass assessment in the ICU.

4.3 Strengths and Limitations of Methods

4.3.1 Functional Assessment

Our analyses were performed in a cohort of 68 previously functionally independent patients from three study populations with ultrasound at the time of ICU admission. The sample size of our study is larger than in many previous studies investigating skeletal muscle ultrasound and its use in predicting functional status. Additionally, we obtained a baseline Mobility-Transfer-Barthel, defined as the level of independence on transfer and mobility on level surfaces two weeks prior to hospital admission. This is a strength of our study, as patients' prior functional capacity has often not been accounted for in studies investigating ultrasound in the ICU. Critical care physicians must often rely on information from proxies regarding prior disability. Therefore, the use of tools to define pre-admission status, such as the Clinical Frailty Scale, as a predictor of outcomes has gained increasing interest (Church, Rogers et al. 2020). Our previously functionally independent cohort certainly does not reflect the heterogenous pre-admission status of typical critically ill populations. Yet, the strong functional decline to discharge observed is a testament to both the illnesses warranting admission and the burden critical care poses on patients. Importantly, nearly half of the patients (31/68) in our study were admitted for traumatic or non-traumatic brain injury. Although sub-analyses by admission diagnosis were not performed, the possibility of traumatic or non-traumatic brain injury as a main driving force of functional decline must be considered. In a study comparing the 20-point Barthel-Index to the Functional Independence Measure (FIM) in 259 young patients admitted to a neurological rehabilitation facility, the mean admission Barthel score was 9/20 and 10/20 several months following non-traumatic and traumatic brain injury, respectively (Houlden, Edwards et al. 2006). This illustrates that even several months following brain injury, these conditions are often associated with serious disability. Although we did not follow patients long-term after discharge, survivors presumably had steep climbs to recovery to baseline status.

While our primary endpoint was functional independence in mobilization at discharge, specifically for the domains transfer and ambulation on level surfaces, we did not perform common muscle strength tests. We chose to evaluate these domains because they are essential to patients'

independence in performing activities such as dressing or bathing. The lack of assessment of further domains essential to patients' independence at discharge is a limitation of our study.

Another important aspect is the timing assessments of functional capacity are performed. While other studies have performed assessments during ICU stay or at ICU discharge, we decided to test the predictive capability of ultrasound for functional capacity at hospital discharge. At this time point, patients are free of intensive care treatments which are impediments to functional assessment. However, during post-discharge rehabilitation further changes in functional capacity are likely. Thus, the ideal time point to test patients' functional capacity remains unknown.

4.3.2 Ultrasound Methods

We aimed to compare the measurement of rectus femoris cross-sectional area with that of muscle thickness to assess muscle loss over the course of critical illness. This is a strength in our methods because most studies have chosen to investigate the change in only one of these measurements using ultrasound. It remains unclear which ultrasound measurement is the better surrogate of true muscle loss. Only two studies compared either of the two measurements with the gold standard method of muscle biopsy. The authors found that thickness measurements underestimated true muscle loss, while rectus femoris cross-sectional area did not (Puthucheary, McNelly et al. 2017) (Puthucheary, Rawal et al. 2013).

A limitation in our ultrasound methods is the discrepancy in measurement protocols among our three study cohorts; 29 of the 282 total ultrasound measurement values were taken at 2/3 and the remainder at 3/5 distance from the anterior superior iliac spine to the patella. However, we believe this did not impact our results, since we did not find a correlation between absolute change on ultrasound and functional capacity at discharge. This reiterates the need for standardized ultrasound measurement protocols to assess skeletal muscle in the ICU. We consider using maximum compression of the thigh with the ultrasound probe as a methodological strength of our study. Many studies measuring muscle thickness have avoided compression due to concerns of potential variability in muscle size and shape (Mourtzakis, Parry et al. 2017). Others have argued that maximum compression eliminates potential confounding of measurements by edema (Paris, Mourtzakis et al. 2017). One study conducted by Ozdemir et al. investigated the effects of

maximum vs. no compression on thickness measurements of the quadriceps muscle in 55 ICU patients. Patients were divided into either a hyper- or euvolemic group based on the size and collapsibility of the inferior vena cava on ultrasound and total fluid balance. The authors found that when using no compression, measurements significantly differed between hyper- and euvolemic patients. On the other hand, they found no significant difference in measurements between groups when using maximum compression (Ozdemir, Ozdemir et al. 2019). This points to the possibility that maximally compressed thickness measurements may be less affected by patients' fluid status. Further studies are needed to better understand the effect of fluid balance on ultrasound muscle measurements. Lastly, the variance between left and right leg values among our cross-sectional area and thickness measurements highlights a degree of imprecision potentially limiting our data.

4.4 Outlook and Future Use

Future research must aim to translate results into simple protocols for use by clinicians given that it remains unknown at which time point, with which method, and with what frequency ultrasound assessment is best applied. A single cut-off value to define low admission muscle mass, which may warrant further monitoring or early interventions to combat muscle wasting is needed. Additional factors that may indicate longstanding muscle wasting prior to admission, baseline functional status, as well as risk factors such as frailty should guide potential intervention. Certain populations of critically ill patients may benefit more than others from screening of muscle mass. Ultrasound has proven to be a quick, easy-to-use, and reliable tool. Larger studies using standardized and comparable ultrasound protocols are needed prior to future clinical integration. Lastly, ultrasound will need to provide information regarding muscle mass earlier, with more accuracy, and greater clarity before guiding implementation of therapies.

5. Conclusion

Critically ill patients lose muscle mass early into ICU stay. It remains unclear how this early muscle wasting is associated with the disability survivors of critical illness experience at hospital discharge. Ultrasound can be used as a quick, reliable, and non-invasive tool to quantify skeletal muscle mass at the bedside. Cross-sectional area of the rectus femoris and thickness of the quadriceps muscle are commonly used measurements. It remains unknown which of these two measurements is the better surrogate of whole-body muscle mass, which is more suited for continuous monitoring during hospital stay, and which can best predict functional status at discharge. We aimed to investigate if skeletal muscle ultrasound cross-sectional area and thickness measurements or their changes during ICU stay correlate with the level of functional independence at hospital discharge. Additionally, we aimed to compare these ultrasound measurements with measurements of CT skeletal muscle area. Almost all patients showed a decline in functional independence over ICU and hospital stay. We found no correlation between ultrasound measurements (at ICU admission and change from ICU admission to ICU discharge) and level of functional independence. Ultrasound cross-sectional area measurements showed a stronger correlation with CT skeletal muscle area than with thickness measurements. In summary, quadriceps ultrasound is an easy and reliable method to quantify skeletal muscle loss. Computer tomography is an accurate tool to quantify skeletal muscle mass, however application in critically ill patients poses challenges. Without standardization of ultrasound measurement protocols across studies, the question remains when and how clinicians should use muscle loss detected via ultrasound to guide preventive measures and counter intensive care induced disability.

6. References

Abe, T., J. P. Loenneke, R. S. Thiebaud and T. Fukunaga (2014). "Age-related site-specific muscle wasting of upper and lower extremities and trunk in Japanese men and women." Age (Dordr) **36**(2): 813-821.
Abe, T., R. S. Thiebaud, J. P. Loenneke, M. Loftin and T. Fukunaga (2014). "Prevalence of site-specific thigh sarcopenia in Japanese men and women." Age (Dordr) **36**(1): 417-426.
Ali, N. A., J. M. O'Brien, Jr., S. P. Hoffmann, G. Phillips, A. Garland, J. C. Finley, K. Almoosa, R. Hejal, K. M. Wolf, S. Lemeshow, A. F. Connors, Jr., C. B. Marsh and C. Midwest Critical Care (2008). "Acquired weakness, handgrip strength, and mortality in critically ill patients." Am J Respir Crit Care Med **178**(3): 261-268.
Annetta, M. G., M. Pittiruti, D. Silvestri, D. L. Grieco, A. Maccaglia, M. F. La Torre, N. Magarelli, G. Mercurio, A. Caricato and M. Antonelli (2017). "Ultrasound assessment of rectus femoris and anterior tibialis muscles in young trauma patients." Ann Intensive Care **7**(1): 104.
Baldwin CE, B. A. (2014). "Alterations in respiratory and limb muscle strength and size in patients with sepsis who are mechanically ventilated." Phys Ther. **Jan;94**(1): 68-82.
Batt, J., M. S. Herridge and C. C. Dos Santos (2019). "From skeletal muscle weakness to functional outcomes following critical illness: a translational biology perspective." Thorax **74**(11): 1091-1098.
Bury, C., R. DeChicco, D. Nowak, R. Lopez, L. He, S. Jacob, D. F. Kirby, N. Rahman and G. Cresci (2021). "Use of Bedside Ultrasound to Assess Muscle Changes in the Critically Ill Surgical Patient." JPEN J Parenter Enteral Nutr **45**(2): 394-402.
Charlson ME, P. P., Ales KL, MacKenzie CR (1987). "A new method of classifying prognostic comorbidity in longitudinal studies: development and validation." J Chron Dis **40**(5): 373-383.
Chen, S., S. Wang, L. Xuan, H. Lu, Z. Hu, C. Zhang and H. Zhang (2018). "Comparison of efficacy and safety between electroacupuncture at 'four sacral points' and conventional electroacupuncture for the treatment of urinary incontinence after stroke: study protocol for a randomised controlled trial." BMJ Open **8**(11): e021783.
Church, S., E. Rogers, K. Rockwood and O. Theou (2020). "A scoping review of the Clinical Frailty Scale." BMC Geriatr **20**(1): 393.
Cruz-Jentoft, A. J., G. Bahat, J. Bauer, Y. Boirie, O. Bruyere, T. Cederholm, C. Cooper, F. Landi, Y. Rolland, A. A. Sayer, S. M. Schneider, C. C. Sieber, E. Topinkova, M. Vandewoude, M. Visser, M. Zamboni, P. Writing Group for the European Working Group on Sarcopenia in Older and E. the Extended Group for (2019). "Sarcopenia: revised European consensus on definition and diagnosis." Age Ageing **48**(1): 16-31.
De Jonghe, B., T. Sharshar, J. P. Lefaucheur, F. J. Authier, I. Durand-Zaleski, M. Boussarsar, C. Cerf, E. Renaud, F. Mesrati, J. Carlet, J. C. Raphaël, H. Outin and S. Bastuji-Garin (2002). "Paresis acquired in the intensive care unit: a prospective multicenter study." Jama **288**(22): 2859-2867.
Desai, S. V., T. J. Law and D. M. Needham (2011). "Long-term complications of critical care." Crit Care Med **39**(2): 371-379.
Englesbe, M. J., S. P. Patel, K. He, R. J. Lynch, D. E. Schaubel, C. Harbaugh, S. A. Holcombe, S. C. Wang, D. L. Segev and C. J. Sonnenday (2010). "Sarcopenia and mortality after liver transplantation." J Am Coll Surg **211**(2): 271-278.
Fetterplace, K., L. Corlette, Y. A. Abdelhamid, J. J. Presneill, M. T. Paris, D. Stella, M. Mourtzakis, C. MacIsaac and A. M. Deane (2021). "Assessment of muscle mass using ultrasound with minimal versus maximal pressure compared with computed tomography in critically ill adult patients." Aust Crit Care **34**(4): 303-310.
Fischer, A., M. Anwar, A. Hertwig, R. Hahn, M. Pesta, I. Timmermann, T. Siebenrock, K. Liebau and M. Hiesmayr (2020). "Ultrasound method of the USVALID study to measure subcutaneous adipose tissue

and muscle thickness on the thigh and upper arm: An illustrated step-by-step guide." Clinical Nutrition Experimental **32**: 38-73.
Gomez-Perez, S. L., J. M. Haus, P. Sheean, B. Patel, W. Mar, V. Chaudhry, L. McKeever and C. Braunschweig (2016). "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 **40**(3): 308-318.
Guerri, S., D. Mercatelli, M. P. Aparisi Gomez, A. Napoli, G. Battista, G. Guglielmi and A. Bazzocchi (2018). "Quantitative imaging techniques for the assessment of osteoporosis and sarcopenia." Quant Imaging Med Surg **8**(1): 60-85.
Hadda, V., R. Kumar, G. C. Khilnani, M. Kalaivani, K. Madan, P. Tiwari, S. Mittal, A. Mohan, A. S. Bhalla and R. Guleria (2018). "Trends of loss of peripheral muscle thickness on ultrasonography and its relationship with outcomes among patients with sepsis." J Intensive Care **6**: 81.
Hermans, G. and G. Van den Berghe (2015). "Clinical review: intensive care unit acquired weakness." Crit Care **19**: 274.
Herridge, M. S., L. M. Chu, A. Matte, G. Tomlinson, L. Chan, C. Thomas, J. O. Friedrich, S. Mehta, F. Lamontagne, M. Levasseur, N. D. Ferguson, N. K. Adhikari, J. C. Rudkowski, H. Meggison, Y. Skrobik, J. Flannery, M. Bayley, J. Batt, C. D. Santos, S. E. Abbey, A. Tan, V. Lo, S. Mathur, M. Parotto, D. Morris, L. Flockhart, E. Fan, C. M. Lee, M. E. Wilcox, N. Ayas, K. Choong, R. Fowler, D. C. Scales, T. Sinuff, B. H. Cuthbertson, L. Rose, P. Robles, S. Burns, M. Cypel, L. Singer, C. Chaparro, C. W. Chow, S. Keshavjee, L. Brochard, P. Hebert, A. S. Slutsky, J. C. Marshall, D. Cook, J. I. Cameron, R. P. Investigators and G. Canadian Critical Care Trials (2016). "The RECOVER Program: Disability Risk Groups and 1-Year Outcome after 7 or More Days of Mechanical Ventilation." Am J Respir Crit Care Med **194**(7): 831-844.
Herridge, M. S., C. M. Tansey, A. Matté, G. Tomlinson, N. Diaz-Granados, A. Cooper, C. B. Guest, C. D. Mazer, S. Mehta, T. E. Stewart, P. Kudlow, D. Cook, A. S. Slutsky and A. M. Cheung (2011). "Functional disability 5 years after acute respiratory distress syndrome." N Engl J Med **364**(14): 1293-1304.
Heymsfield, S. B., M. C. Gonzalez, J. Lu, G. Jia and J. Zheng (2015). "Skeletal muscle mass and quality: evolution of modern measurement concepts in the context of sarcopenia." Proc Nutr Soc **74**(4): 355-366.
Houlden, H., M. Edwards, J. McNeil and R. Greenwood (2006). "Use of the Barthel Index and the Functional Independence Measure during early inpatient rehabilitation after single incident brain injury." Clin Rehabil **20**(2): 153-159.
Iwashyna, T. J., E. W. Ely, D. M. Smith and K. M. Langa (2010). "Long-term cognitive impairment and functional disability among survivors of severe sepsis." JAMA **304**(16): 1787-1794.
Joskova, V., A. Patkova, E. Havel, S. Najpaverova, D. Uramova, M. Kovarik, Z. Zadak and M. Hronek (2018). "Critical evaluation of muscle mass loss as a prognostic marker of morbidity in critically ill patients and methods for its determination." J Rehabil Med **50**(8): 696-704.
Katari, Y., R. Srinivasan, P. Arvind and S. Hiremathada (2018). "Point-of-Care Ultrasound to Evaluate Thickness of Rectus Femoris, Vastus Intermedius Muscle, and Fat as an Indicator of Muscle and Fat Wasting in Critically Ill Patients in a Multidisciplinary Intensive Care Unit." Indian J Crit Care Med **22**(11): 781-788.
Kleyweg, R. P., F. G. A. Van Der Meché and P. I. M. Schmitz (1991). "Interobserver agreement in the assessment of muscle strength and functional abilities in Guillain-Barré syndrome." Muscle & Nerve **14**(11): 1103-1109.
Knaus WA, D. E., Wagner DP, Zimmerman JE (1985). "APACHE II: a severity of disease classification system." Crit Care Med **13**(10): 818-829.
Kress, J. P. and J. B. Hall (2014). "ICU-acquired weakness and recovery from critical illness." N Engl J Med **370**(17): 1626-1635.

Lad, H., T. M. Saumur, M. S. Herridge, C. C. Dos Santos, S. Mathur, J. Batt and P. M. Gilbert (2020). "Intensive Care Unit-Acquired Weakness: Not just Another Muscle Atrophying Condition." Int J Mol Sci **21**(21).
Lambell, K. J., A. C. Tierney, J. C. Wang, V. Nanjayya, A. Forsyth, G. S. Goh, D. Vicendese, E. J. Ridley, S. M. Parry, M. Mourtzakis and S. J. King (2021). "Comparison of Ultrasound-Derived Muscle Thickness With Computed Tomography Muscle Cross-Sectional Area on Admission to the Intensive Care Unit: A Pilot Cross-Sectional Study." JPEN J Parenter Enteral Nutr **45**(1): 136-145.
Lamontagne, F., B. Rochwerg, L. Lytvyn, G. Guyatt, M. Møller, D. Annane, M. Kho, N. Adhikari, F. Machado, P. Vandvik, P. Dodek, R. Leboeuf, M. Briel, M. Hashmi, J. Camsooksai, M. Shankar-Hari, M. Baraki, K. Fugate, S. Chua and R. Siemieniuk (2018). "Corticosteroid therapy for sepsis: A clinical practice guideline." BMJ **362**: k3284.
Looijaard, W., J. Molinger and P. J. M. Weijs (2018). "Measuring and monitoring lean body mass in critical illness." Curr Opin Crit Care **24**(4): 241-247.
Loosen, S. H., M. Schulze-Hagen, T. Pungel, L. Bundgens, T. Wirtz, J. N. Kather, M. Vucur, P. Paffenholz, M. Demir, P. Bruners, C. Kuhl, C. Trautwein, F. Tacke, T. Luedde, A. Koch and C. Roderburg (2020). "Skeletal Muscle Composition Predicts Outcome in Critically Ill Patients." Crit Care Explor **2**(8): e0171.
Mayer, K. P., M. L. Thompson Bastin, A. A. Montgomery-Yates, A. M. Pastva, E. E. Dupont-Versteegden, S. M. Parry and P. E. Morris (2020). "Acute skeletal muscle wasting and dysfunction predict physical disability at hospital discharge in patients with critical illness." Crit Care **24**(1): 637.
Moisey, L. L., M. Mourtzakis, B. A. Cotton, T. Premji, D. K. Heyland, C. E. Wade, E. Bulger and R. A. Kozar (2013). "Skeletal muscle predicts ventilator-free days, ICU-free days, and mortality in elderly ICU patients." Crit Care **17**(5): R206.
Mourtzakis, M., S. Parry, B. Connolly and Z. Puthucheary (2017). "Skeletal Muscle Ultrasound in Critical Care: A Tool in Need of Translation." Ann Am Thorac Soc **14**(10): 1495-1503.
Mourtzakis, M., C. M. Prado, J. R. Lieffers, T. Reiman, L. J. McCargar and V. E. Baracos (2008). "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 **33**(5): 997-1006.
Mueller, N., S. Murthy, C. R. Tainter, J. Lee, K. Riddell, F. J. Fintelmann, S. D. Grabitz, F. P. Timm, B. Levi, T. Kurth and M. Eikermann (2016). "Can Sarcopenia Quantified by Ultrasound of the Rectus Femoris Muscle Predict Adverse Outcome of Surgical Intensive Care Unit Patients as well as Frailty? A Prospective, Observational Cohort Study." Ann Surg **264**(6): 1116-1124.
Ng, C. C., Z. Y. Lee, W. Y. Chan, M. F. Jamaluddin, L. J. Tan, P. N. Sitaram, S. R. Ruslan and M. S. Hasan (2020). "Low Muscularity as Assessed by Abdominal Computed Tomography on Intensive Care Unit Admission Is Associated With Mortality in a Critically Ill Asian Population." JPEN J Parenter Enteral Nutr **44**(3): 425-433.
Oeyen, S. G., D. M. Vandijck, D. D. Benoit, L. Annemans and J. M. Decruyenaere (2010). "Quality of life after intensive care: a systematic review of the literature." Crit Care Med **38**(12): 2386-2400.
Ozdemir, U., M. Ozdemir, G. Aygencel, B. Kaya and M. Turkoglu (2019). "The role of maximum compressed thickness of the quadriceps femoris muscle measured by ultrasonography in assessing nutritional risk in critically-ill patients with different volume statuses." Rev Assoc Med Bras (1992) **65**(7): 952-958.
Palakshappa, J. A., J. P. Reilly, W. D. Schweickert, B. J. Anderson, V. Khoury, M. G. Shashaty, D. Fitzgerald, C. Forker, K. Butler, C. A. Ittner, R. Feng, D. C. Files, M. P. Bonk, J. D. Christie and N. J. Meyer (2018). "Quantitative peripheral muscle ultrasound in sepsis: Muscle area superior to thickness." J Crit Care **47**: 324-330.
Paris, M. T., M. Mourtzakis, A. Day, R. Leung, S. Watharkar, R. Kozar, C. Earthman, A. Kuchnia, R. Dhaliwal, L. Moisey, C. Compher, N. Martin, M. Nicolo, T. White, H. Roosevelt, S. Peterson and D. K.

Heyland (2017). "Validation of Bedside Ultrasound of Muscle Layer Thickness of the Quadriceps in the Critically Ill Patient (VALIDUM Study)." JPEN J Parenter Enteral Nutr **41**(2): 171-180.
Parry, S. M., D. El-Ansary, M. S. Cartwright, A. Sarwal, S. Berney, R. Koopman, R. Annoni, Z. Puthucheary, I. R. Gordon, P. E. Morris and L. Denehy (2015). "Ultrasonography in the intensive care setting can be used to detect changes in the quality and quantity of muscle and is related to muscle strength and function." J Crit Care **30**(5): 1151 e1159-1114.
Pasta, G., G. Nanni, L. Molini and S. Bianchi (2010). "Sonography of the quadriceps muscle: Examination technique, normal anatomy, and traumatic lesions." J Ultrasound **13**(2): 76-84.
Perkisas, S., S. Baudry, J. Bauer, D. Beckwée, A.-M. De Cock, H. Hobbelen, H. Jager-Wittenaar, A. Kasiukiewicz, F. Landi, E. Marco, A. Merello, K. Piotrowicz, E. Sanchez, D. Sanchez-Rodriguez, A. Scafoglieri, A. Cruz-Jentoft and M. Vandewoude (2018). "Application of ultrasound for muscle assessment in sarcopenia: towards standardized measurements." European Geriatric Medicine **9**(6): 739-757.
Pistoia, F., S. Sacco, M. Sara and A. Carolei (2013). "The perception of pain and its management in disorders of consciousness." Curr Pain Headache Rep **17**(11): 374.
Prado, C. M. M., J. R. Lieffers, L. J. McCargar, T. Reiman, M. B. Sawyer, L. Martin and V. E. Baracos (2008). "Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study." The Lancet Oncology **9**(7): 629-635.
Puthucheary, Z. A., A. S. McNelly, J. Rawal, B. Connolly, P. S. Sidhu, A. Rowlerson, J. Moxham, S. D. Harridge, N. Hart and H. E. Montgomery (2017). "Rectus Femoris Cross-Sectional Area and Muscle Layer Thickness: Comparative Markers of Muscle Wasting and Weakness." Am J Respir Crit Care Med **195**(1): 136-138.
Puthucheary, Z. A., J. Rawal, M. McPhail, B. Connolly, G. Ratnayake, P. Chan, N. S. Hopkinson, R. Phadke, T. Dew, P. S. Sidhu, C. Velloso, J. Seymour, C. C. Agley, A. Selby, M. Limb, L. M. Edwards, K. Smith, A. Rowlerson, M. J. Rennie, J. Moxham, S. D. Harridge, N. Hart and H. E. Montgomery (2013). "Acute skeletal muscle wasting in critical illness." JAMA **310**(15): 1591-1600.
Rawal, G., S. Yadav and R. Kumar (2017). "Post-intensive Care Syndrome: an Overview." J Transl Int Med **5**(2): 90-92.
Rockwood, K., X. Song, C. MacKnight, H. Bergman, D. B. Hogan, I. McDowell and A. Mitnitski (2005). "A global clinical measure of fitness and frailty in elderly people." Cmaj **173**(5): 489-495.
Rydingsward, J. E., C. M. Horkan, K. M. Mogensen, S. A. Quraishi, K. Amrein and K. B. Christopher (2016). "Functional Status in ICU Survivors and Out of Hospital Outcomes: A Cohort Study." Crit Care Med **44**(5): 869-879.
Sheean, P. M., S. J. Peterson, S. Gomez Perez, K. L. Troy, A. Patel, J. S. Sclamberg, F. C. Ajanaku and C. A. Braunschweig (2014). "The prevalence of sarcopenia in patients with respiratory failure classified as normally nourished using computed tomography and subjective global assessment." JPEN J Parenter Enteral Nutr **38**(7): 873-879.
Shen, W., M. Punyanitya, Z. Wang, D. Gallagher, M. P. St-Onge, J. Albu, S. B. Heymsfield and S. Heshka (2004). "Total body skeletal muscle and adipose tissue volumes: estimation from a single abdominal cross-sectional image." J Appl Physiol (1985) **97**(6): 2333-2338.
Skinner, E. H., S. Berney, S. Warrillow and L. Denehy (2009). "Development of a physical function outcome measure (PFIT) and a pilot exercise training protocol for use in intensive care." Crit Care Resusc **11**(2): 110-115.
Subbe, C. P., C. Burford, I. Le Jeune, C. Masterton-Smith and D. Ward (2015). "Relationship between input and output in acute medicine - secondary analysis of the Society for Acute Medicine's benchmarking audit 2013 (SAMBA '13)." Clin Med (Lond) **15**(1): 15-19.
Teasdale, G. and B. Jennett (1974). "Assessment of coma and impaired consciousness. A practical scale." Lancet **2**(7872): 81-84.

Teigen, L. M., A. J. Kuchnia, E. Nagel, C. Deuth, D. M. Vock, U. Mulasi and C. P. Earthman (2018). "Impact of Software Selection and ImageJ Tutorial Corrigendum on Skeletal Muscle Measures at the Third Lumbar Vertebra on Computed Tomography Scans in Clinical Populations." JPEN J Parenter Enteral Nutr **42**(5): 933-941.
Thomaes, T., M. Thomis, S. Onkelinx, W. Coudyzer, V. Cornelissen and L. Vanhees (2012). "Reliability and validity of the ultrasound technique to measure the rectus femoris muscle diameter in older CAD-patients." BMC Med Imaging **12**: 7.
Ticinesi, A., T. Meschi, M. V. Narici, F. Lauretani and M. Maggio (2017). "Muscle Ultrasound and Sarcopenia in Older Individuals: A Clinical Perspective." J Am Med Dir Assoc **18**(4): 290-300.
Tillquist, M., D. J. Kutsogiannis, P. E. Wischmeyer, C. Kummerlen, R. Leung, D. Stollery, C. J. Karvellas, J. C. Preiser, N. Bird, R. Kozar and D. K. Heyland (2014). "Bedside ultrasound is a practical and reliable measurement tool for assessing quadriceps muscle layer thickness." JPEN J Parenter Enteral Nutr **38**(7): 886-890.
van Ruijven, I. M., S. N. Stapel, J. Molinger and P. J. M. Weijs (2021). "Monitoring muscle mass using ultrasound: a key role in critical care." Curr Opin Crit Care **27**(4): 354-360.
Vincent JL, M. R., Takala J, Willatts S, De Mendonça A, Bruining H, Reinhart CK, Suter PM, Thijs LG (1996). "The SOFA (Sepsis-related Organ Failure Assessment) score to describe organ dysfunction/failure. On behalf of the Working Group on Sepsis-Related Problems of the European Society of Intensive Care Medicine." Intensive Care Med **7**(22): 707-710.
Weinel, L. M., M. J. Summers and L. A. Chapple (2019). "Ultrasonography to measure quadriceps muscle in critically ill patients: A literature review of reported methodologies." Anaesth Intensive Care **47**(5): 423-434.
Wilkinson, T. J., E. F. Gore, N. Vadaszy, D. G. D. Nixon, E. L. Watson and A. C. Smith (2021). "Utility of Ultrasound as a Valid and Accurate Diagnostic Tool for Sarcopenia: Sex-Specific Cutoff Values in Chronic Kidney Disease." J Ultrasound Med **40**(3): 457-467.
Yang, H., Y. H. Chen, T. F. Hsieh, S. Y. Chuang and M. J. Wu (2016). "Prediction of Mortality in Incident Hemodialysis Patients: A Validation and Comparison of CHADS2, CHA2DS2, and CCI Scores." PLoS One **11**(5): e0154627.

7. Appendix

Glasgow Coma Scale. Modified after Pistoia, Sacco et al. 2013 (Pistoia, Sacco et al. 2013).

Score	response
Eye opening	
4	Opens eyes spontaneously
3	Opens eyes in response to speech
2	*Opens eyes in response to painful stimulations*
1	Does not open eyes in response to any stimulation
Motor response	
6	Follows commands
5	*Makes localized movement in response to painful stimulation*
4	*Makes non purposeful movements in response to painful stimulation* (withdraws from pain)
3	*Flexes upper extremities / extends lower extremities in response to painful stimulation*
2	*Extends all extremities in response to painful stimulation*
1	*Makes no response to noxious stimuli*
Verbal response	
5	Is oriented to person, place and time
4	Converses, may be confused
3	Replies with inappropriate words
2	Makes incomprehensible sounds
1	Makes no response

Items involving painful stimulations are rendered in italics

Charlson Comorbidity Index. Modified after Yang, Chen et al. 2016 (Yang, Chen et al. 2016).

Comorbidity	Score
Prior myocardial infarction	1
Congestive heart failure	1
Peripheral vascular disease	1
Cerebrovascular disease	1
Dementia	1
Chronic pulmonary disease	1
Rheumatologic disease	1
Peptic ulcer disease	1
Mild liver disease	1
Diabetes	1
Cerebrovascular (hemiplegia) event	2
Moderate-to-severe renal disease	2
Diabetes with chronic complications	2
Cancer without metastases	2
Leukemia	2
Lymphoma	2
Moderate or severe liver disease	3
Metastatic solid tumor	6
Acquired immuno-deficiency syndrome (AIDS)	6

doi:10.1371/journal.pone.0154627.t003

Clinical Frailty Scale. Modified after Subbe, Burford et al. 2015 (Subbe, Burford et al. 2015).

Clinical Frailty Scale*

1 Very Fit – People who are robust, active, energetic and motivated. These people commonly exercise regularly. They are among the fittest for their age.

2 Well – People who have **no active disease symptoms** but are less fit than category 1. Often, they exercise or are very **active occasionally**, e.g. seasonally.

3 Managing Well – People whose **medical problems are well controlled,** but are **not regularly active** beyond routine walking.

4 Vulnerable – While **not dependent** on others for daily help, often **symptoms limit activities.** A common complaint is being "slowed up", and/or being tired during the day.

5 Mildly Frail – These people often have **more evident slowing,** and need help in **high order IADLs** (finances, transportation, heavy housework, medications). Typically, mild frailty progressively impairs shopping and walking outside alone, meal preparation and housework.

6 Moderately Frail – People need help with **all outside activities** and with **keeping house.** Inside, they often have problems with stairs and need **help with bathing** and might need minimal assistance (cuing, standby) with dressing.

7 Severely Frail – **Completely dependent for personal care**, from whatever cause (physical or cognitive). Even so, they seem stable and not at high risk of dying (within ~ 6 months).

8 Very Severely Frail – Completely dependent, approaching the end of life. Typically, they could not recover even from a minor illness.

9 Terminally Ill - Approaching the end of life. This category applies to people with **a life expectancy <6 months**, who are **not otherwise evidently frail.**

Scoring frailty in people with dementia

The degree of frailty corresponds to the degree of dementia. Common **symptoms in mild dementia** include forgetting the details of a recent event, though still remembering the event itself, repeating the same question/story and social withdrawal.

In **moderate dementia**, recent memory is very impaired, even though they seemingly can remember their past life events well. They can do personal care with prompting.

In **severe dementia**, they cannot do personal care without help.

* 1. Canadian Study on Health & Aging, Revised 2008.
2. K. Rockwood et al. A global clinical measure of fitness and frailty in elderly people. CMAJ 2005;173:489-495.

APACHE II Severity of Disease Classification System. Modified after Knaus et al. 1985 (Knaus WA 1985).

The APACHE II Severity of Disease Classification System

Physiologic Variable	+4	+3	+2	+1	0	+1	+2	+3	+4
Temperature - rectal (°C)	≥41	39-40.9		38.5-38.9	36-38.4	34-35.9	32-33.9	30-31.9	≤29.9
Mean Arterial Pressure (mm Hg)	≥160	130-159	110-129		70-109		50-69		≤49
Heart Rate	≥180	140-179	110-139		70-109		55-69	40-54	≤39
Respiratory Rate (nonventilated or ventilated)	≥50	35-49		25-34	12-24	10-11	6-9		≤5
Oxygenation (mmHg) a. $FiO_2 > 0,5$ use $A\text{-}aDO_2$ b. $FiO_2 < 0,5$ use PaO_2	a ≥500	350-499	200-349		<200				
	b				> 70	61-70		55-60	<55
Arterial pH	≥7.7	7.6-7.69		7.5-7.59	7.33-7.49		7.25-7.32	7.15-7.24	<7.15
Serum Sodium (mmol/l)	≥180	160-179	155-159	150-154	130-149		120-129	111-119	≤110
Serum Potassium (mmol/l)	≥7	6-6.9		5.5-5.9	3.5-5.4	3-3.4	2.5-2.9		<2.5
Serum Creatinine (mg/dl, Double point score for acute renal failure)	≥3.5	2-3.4	1.5-1.9		0.6-1.4		<0.6		
Hematocrit (%)	≥60		50-59.9	46-49.9	30-45.9		20-29.9		<20
White Blood Count (in 1000/mm³)	≥40		20-39.9	15-19.9	3-14.9		1-2.9		<1
Glasgow-Coma-Scale (GCS)	Score = 15 minus actual GCS								
Serum HCO_3 (venous, mmol/l, use if no ABGs)	≥52	41-51.9		32-40.9	22-31.9		18-21.9	15-17.9	<15
A = Total Acute Physiology Score APS	Sum of the 12 individual variable points								

B = Age Points

≤44 years	0 points
45-54 years	2 points
55-64 years	3 points
65-74 years	5 points
≥75 years	6 points

C = Chronic Health Points

If the patient has a history of severe organ system insufficiency or is immunocompromised assign points as follows:

a. For nonoperative or emergency postoperative patients – 5 points

b. For elective postoperative patients – 2 points

APACHE II Score = Sum of A (APS points) + B (Age points) + C (Chronic Health points)

(From: Knaus WA, Draper EA, Wagner DP, Zimmerman JE. APACHE II: a severity of disease classification system. Crit Care Med 1985;13(10):818-29)

Sequential Organ Failure Assessment Score. Modified after Lamontagne, Rochwerg et al. 2018 (Lamontagne, Rochwerg et al. 2018).

System or organ and measure	SOFA score				
	0	1	2	3	4
Respiratory:					
P_aO_2/FiO_2, mm Hg	≥400	300-399	200-299	100-199 with respiratory support	<100 with respiratory support
Coagulation:					
Platelets, × 10^3/µL	≥150	100-149	50-99	20-49	<20
Liver:					
Bilirubin, µmol/L (mg/dL)	<20 (1.2)	20-32 (1.2-1.9)	33-101 (2.0-5.9)	102-204 (6.0-11.9)	>204 (12.0)
Circulatory:					
Mean arterial pressure, mm Hg	≥70	<70	Low dose dopamine or any dose dobutamine	Low-medium dose noradrenalin or adrenalin; medium dose dopamine	High dose noradrenalin, adrenalin, or dopamine
Central nervous system:					
Glasgow Coma Scale score	15	13-14	10-12	6-9	<6
Renal:					
Creatinine, µmol/L (mg/dL)	<110 (1.2)	110-170 (1.2-1.9)	171-299 (2.0-3.4)	300-440 (3.5-4.9)	>440 (5.0)
Urine output, mL/day	–	–	–	<500	<200

*Our recommendation applies to patients with an infection and a SOFA score of ≥2.
P_aO_2 = partial pressure of oxygen (arterial). F_iO_2 = fraction of inspired oxygen.

Barthel Activities of Daily Living Index. Mobility points were added for categories “Transfer” and “Mobility”, totaling 30 maximum points as the endpoint for functional independence. Modified after Chen, Wang et al. 2018 (Chen, Wang et al. 2018).

The Barthel Index		Patient Name Rater Name Date:
Activity		**Score**
Feeding	Unable	0
	Some help required (eg, needs help cutting, spreading butter, etc. or requires a modified diet)	5
	Independent	10
Bathing	Dependent	0
	Independent (or in shower)	5
Grooming	Needs help with personal care	0
	Independent face/hair/teeth/shaving (implements provided)	5
Dressing	Dependent	0
	Needs help but can do at least half unaided	5
	Independent (including buttons, zips, laces, etc.)	10
Bowels	Incontinent or catheterized and unable to manage alone	0
	Occasional accident	5
	Continent	10
Bladder	Incontinent or catheterized and unable to manage alone	0
	Occasional accident	5
	Continent	10
Toilet use	Dependent	0
	Needs some help, but can do some things alone	5
	Independent (can get on and off, dress and wipe unassisted)	10
Transfer (bed to chair and back)	Unable, no sitting balance	0
	Major help (one or two people, physical), can sit	5
	Minor help (verbal or physical)	10
	Independent	15
Mobility (on level surfaces)	Immobile or <50 yards	0
	Wheelchair independent, including corners; >50 yards	5
	Walks with little help from one person (verbal or physical); >50 yards	10
	Independent (but may use an aid; for example, walking stick); >50 yards	15
Stairs	Unable	0
	Needs help (verbal, carrying aid)	5
	Independent	10
	Total	