Reliability and feasibility of skeletal muscle ultrasound in the acute burn setting

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TITLE PAGE

Reliability and feasibility of skeletal muscle ultrasound in the acute burn setting.

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DECLARATION OF INTEREST

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MANUSCRIPT

INTRODUCTION

Ultrasound is widely used to measure changes in musculature in various disease populations, however, not in burn patients[1]. This is noteworthy since the persistent loss of muscle mass following burns is a well-documented metabolic phenomenon with tremendous potential to impact short and long-term health[2–7]. Burn injury is characterised by a sustained hypermetabolic response (i.e. elevated energy expenditure) which induces significant muscle wasting[10]. In the presence of burn trauma, muscle mass is thought to act as a gradually depleting functional reserve pool providing fuel for vital processes involved in the immune response and wound healing[8–10]. Prolonged periods of inactivity and the administration of corticosteroids, neuromuscular blockers, sedatives, and inadequate nutrition further exacerbate skeletal muscle wasting[11–14]. The resulting loss of muscle mass negatively impacts recovery by, among others, delaying wound healing, increasing infection rates, and prolonging time on mechanical ventilation[15–17]. These clinical complications create a chain reaction wherein rehabilitation is delayed and ICU and hospital length of stay is protracted, ultimately leading to higher in-hospital morbidity, mortality, and health care expenses[17,18]. Beyond these short-term outcomes, muscle wasting is also a likely key aetiological factor in the observed increased risk of burn survivors to develop long-term metabolic, cardiovascular and musculoskeletal disorders[19–28].

Preserving muscle mass is recommended as a therapeutic goal in burn care[29,30]. However, muscle wasting is not generally measured in burn centres[1], likely due to the lack of practical and accurate tools capable of monitoring muscle mass at the bedside. In the absence of such tools, the assessment of muscle mass has commonly been substituted with the assessment of muscle function[1]. However, as muscle plays a pivotal role in the metabolic response to burns it should also be measured independently of its musculoskeletal function[31]. Moreover, the assessment of muscle function, by e.g. force measurements, is often not possible in the acute care setting where sedation and pain may limit patient cooperation.

Muscle ultrasound has been used in the wider critically-ill population as a practical and affordable surrogate measure of whole-body muscle mass at the bedside[38,39]. Its benefits comprise its low costs, the absence of radiation exposure, and its availability as a standard equipment in most burn intensive care units[40]. Particularly, when measured at the level of the quadriceps muscle, it has shown to provide reliable and valid information on the evolution of muscle architecture[41–50]. Whether this can be extrapolated to the burn-injured patient currently remains unknown. Open wounds, varying fluid status, and the limited time window during which dressings are removed are amongst factors that pose significant challenges to the use of ultrasound in this patient population. Its use is further complicated by the presence of multiple methodologies used to obtain ultrasound-derived parameters of the quadriceps muscle[51]. The location of the measurement, the amount of tissue compression, and whether to use an average or a single measurement remain unanswered questions.

Therefore, the present study was initiated to examine the reliability and feasibility of quadriceps muscle ultrasound measures in the acute burn setting. As such, it will lay the groundwork to determine if and how muscle ultrasound could help clinicians and researchers to better identify, treat and monitor burn patients with altered muscle mass.

MATERIAL AND METHODS

This multi-centre cross-sectional reliability and feasibility study was approved by the institutional review board of the Ziekenhuis Netwerken Antwerpen (5018) and the Universitair Ziekenhuis Antwerpen on (B300201942189).

Study population

Twenty adults with burns were recruited between May 2020 and April 2021 upon admission to two Belgian burn centres (ZNA Stuivenberg & Military Hospital Queen Astrid), as part of a larger intervention trial investigating the effects of exercise during the acute phase of burns. Burn subjects were eligible if they met the eligibility criteria as listed in table 1. In addition, this study included twenty healthy adults control subjects to assess how much variability in reliability and feasibility originate from the ultrasound protocol itself versus the subjects in whom it is applied. The healthy subjects were recruited through convenience sampling and assessed in our metabolic research lab (M2RUN) at the University of Antwerp prior to assessing the burn cohort. Assuming a minimum ICC value of 0.75 for interobserver reliability, with alpha 0.05 and 80% power, we estimated a priori that we would need to enrol at least eighteen subjects. Hence, we enrolled twenty subjects to ensure sufficient power. All recruited subjects or their next-of-kin gave informed consent prior to ultrasound assessment.

Measurement protocol:

Two trained assessors (D.R.S. and D.D.) carried out measurements using B-mode ultrasound with a multifrequency linear transducer of either the SonoSite X-porte (FUJIFILM SonoSite, Brussels, Belgium) or the LOGIC V2 and VIVID S5 (GE Healthcare, Machelen, Belgium). Measurements were performed consecutively and in randomised order (random number sequencing generated using Microsoft Excel). Assessors were blinded from each other's measurements. Burn subjects were assessed within 72 hours of burn centre admission by the two assessors to provide feasibility and reliability data at baseline. To provide additional data for feasibility of ultrasound over the course of burn centre stay, one assessor (D.R.S.) carried out up to three follow-up measurements in burn subjects dependent on the burn centre length of stay.

A protocol for the assessment of quadriceps muscle size based on previous work[43,52–54] was adapted to acute burn patients with the help of experts in the field of musculoskeletal ultrasound imaging. Ultrasound-derived muscle parameters were quadriceps muscle layer thickness (QMLT) and rectus femoris cross-sectional area (RF-CSA) as shown in figure 1. QMLT, as commonly defined[52], is equal to the distance between the superior fascial layer of the rectus femoris muscle and the top of the femoral periosteum, comprising the combined thickness of the rectus femoris and intermedius muscle. QMLT was measured at two sites on the anterior aspect of the thigh – at the halfway (referred to as proximal from here on) and two-third (referred to as distal from here on) point of the distance on the midline between the anterior superior iliac spine and the superior patellar pole[52] (figure 2). Image depth was as shallow as possible to visualise the femur, and if necessary, the transducer was tilted in the transversal plane to achieve central position of the femur (figure 1). Two different compression techniques were used for determining QMLT – a maximum and no-compression technique. During maximum-compression the transducer was progressively compressed into the quadriceps muscle until additional pressure did not produce further of QMLT, as previously described[43]. This technique is thought to account for generalised intramuscular oedema which might interfere with measurement of true muscle size[38,43]. For the no-compression technique, the transducer and skin were separated by excess ultrasound transmission gel, avoiding distortion of muscle contour[52,55]. During this manoeuvre the transducer was gradually released until transmission was lost and no more structures were visible. Measurements with the no-compression technique preceded measurements with maximum-compression technique to account for a potential after-effect of compression.

RF-CSA was defined as the surface area within the rectus femoris muscle fascia. RF-CSA was measured at the most proximal point on the midline where the entire muscle contour of the rectus femoris muscle was still visible[53]. The transducer was moved away from the midline (laterally or medially) to ensure positioning directly above the middle of the muscle belly of the rectus femoris. RF-CSA was determined with no-compression only, using the same principles as described for the no-compression technique for determining QMLT (figure 1).

All measurements were carried out with the subject in supine lying with straight knees and hips, and neutral rotation in the hips. If not tolerated by the patients, the head of the bed was elevated up to 30° prior to the measurements[56]. The transducer position remained perpendicular to the midline between the anterior superior iliac spine and the superior patellar pole at each measurement point. On open wound surfaces measurements were carried out using sterile ultrasound gel, sterile probe covers, sterile skin location markings (either by surgical marker or by sterile strips), and sterile measurement tape. In terms of ultrasound-specific parameters, the gain was always kept at zero and the applied depth was set as shallow as possible to visualise structures of interest while ensuring highest resolution. Both thighs were assessed, and each measurement was repeated three times.

Data analysis:

Ultrasound data was stored as anonymised DICOM four to five seconds clips, and frames were selected and analysed using a DICOM reader software (Horos™ viewer v3.3.6, Horos Project) by a blinded assessor who was not present during the data collection. Frames were selected if no further change in muscle contour took place and image quality allowed delineation of structures of interest. For QMLT, a straight line was drawn from the top centre of the ultrasound image towards the middle of femoral shaft. Along this line, a second line was drawn from the inferior border of the superior muscle fascia of the rectus femoris towards the superior border of the femoral periosteum, to determine QMLT (figure 1). For RF-CSA, a closed polygon tool was used to trace the inner lining of the rectus femoris muscle fascia (figure 1).

To determine whether it would be sufficient to use a single measurement as opposed to the average of three measurements, the first of three measurements was used as the single measurement and compared with the average of three measurements.

Differences in demographics between healthy and burn subjects were analysed using two-sample t-tests. Inter-rater reliability of QMLT and RF-CSA was analysed using two-way random effects model with absolute agreement for the calculation of intraclass correlation coefficients (ICC) estimates ICC (2,3) when the average of three measurements was used, or ICC (2,1) for single measurements[57]. Limits of agreement and systematic bias were assessed using Bland Altman plots and one-sample t-tests. Proportional bias was evaluated by means of linear regression. Minimum detectable changes at the 95% confidence interval (MDC95) were calculated with the formula [$MDC95 = SEM*1.96*\sqrt{2}$][58]. The SEM was derived by $SEM = SD \text{ x } \sqrt{(1 - ICC)}$, where the SD represents the pooled standard deviation for the two raters[58]. Normality of data was determined with a one-sample Kolmogorov-Smirnov test. Where necessary, data was log-transformed to meet normality assumptions. However, when the difference between transformed and non-transformed ICCs was less than 1%, we chose to report the ICC and MDC95 on the original scale. ICC estimates and their 95% confidence interval were defined as ICC <0.5 indicative of poor reliability, moderate reliability if between ≥0.5 and <0.75, good reliability if between ≥0.75 and 0.9, and excellent reliability if greater than 0.9[58]. Significance was set at $p < 0.05$. All statistical analysis was performed using SPSS Statistics Version 25 (IBM, USA).

Feasibility was determined by the number of attempted vs. realised measurements, with reasons for measurement failure noted. Finally, the duration of measurements was calculated from the first to the last recorded ultrasound clip for both assessors.

RESULTS

Characteristics

Table 2 shows the characteristics of the recruited burn and healthy subjects. The burn group was comprised of fewer female subjects (n=4) than the healthy group (n=10). Besides this gender differences, groups were comparable, with non-significant differences in age (p=0.11) and body mass index (p=0.54). The total body surface area of burn subjects ranged from 10 to 70%, with nearly half of all measurements taking place on thighs with open wounds. A total of 1971 ultrasound clips from burn subjects (1122 at baseline + 849 at follow-up) and 1200 clips from healthy subjects were collected and analysed over the course of the study period. Sixteen out of twenty burn subjects provided data for follow-up measurements throughout burn centre stay, with four subjects unable to be followed-up for the following reasons: death n=2, repatriation n=1, psychosis n=1. Thirty follow-up measurements took place an average of 5.8 weeks after baseline assessment [95%CI 4.3 – 7.4].

Feasibility

Met opmerkingen [DS1]: @Eric / Ulrike: I created a table about the feasibility (see table 3 after the references). Is this table useful to keep? It was suggested by one reviewer instead of the figure, which I deleted). I describe the most important data in this paragraph, but the table provides more detailed feasibility data for every FU point, and might be good to reference in case they may have more questions about this. What do you think?

The complete measurement procedure in burn subjects of both thighs took an average of 22 min [CI95% 18 – 27] for rater D.R.S. and 20 min [CI95% 16 – 26] for rater D.D. for the baseline assessment, and 22 min for any follow-up measurements, with an overall ratio of approximately 2:1 for QMLT to RF-CSA measurement duration. Using no-compression to measure QMLT deemed feasible for both proximal and distal locations (92.5% and 97.5% of attempted measurements in burn subjects at baseline, 95% and 95% during follow-up measurements, and 95% and 95% in healthy subjects). The reason for measurement failure at baseline were very large thighs (>6 cm depth) due to obesity, high muscularity, oedema, or a combination of these factors, while during follow-up measurements non-penetrable donor site dressings rendered three out of sixty thighs inaccessible. In 10% of proximal and 12.5% distal of attempted measurements in burn subjects at baseline, maximum-compression was not tolerated on open wounds due to pain, whereas on intact skin this was tolerated in all cases. During follow-up measurements, maximum compression was not possible in 8.3% of cases due to pain on open wounds (3.3%) and non-penetrable donor sites (5%). All attempted RF-CSA measurements in healthy and burn subjects at baseline were successfully completed, while during follow-up measurements 5% of cases thighs were non accessible due to non-penetrable donor sites. Of follow-up measurements, all failed attempts took place during the first follow-up measurements (n=16; mean 3.6 weeks after admission assessment, 95%CI 2.4 - 4.9 weeks). All further follow-up assessment at an average of 7 weeks (n = 11) and 14 weeks (n=3), or at discharge (n=16) were 100% feasible for any of the studied parameters. Beyond pain on some open wounds during maximum-compression, no other adverse events occurred.

Reliability

All reliability parameters are shown in table 3 (burn subjects) and table A.1 (healthy subjects). Measurements of QMLT yielded ICC values above 0.9 in all subjects. RF-CSA measurements achieved ICC values ranging from 0.76 to 0.99 in burn subjects and above 0.9 in healthy subjects. MDC95 values ranged between 5 and 18% of the mean score in burn subjects, and 2 and 16% in healthy subjects. Limits of agreement between raters were acceptable for all ultrasound-derived parameters in all subjects (figure 3 and figure A.1). RF-CSA showed somewhat larger limits of agreements than QMLT measurements. In burn subjects, mean differences between raters ranged from 0.01 to 0.1 cm depending on the administered methodology (table 4). In healthy subjects, mean differences between raters were very small, ranging between 0.01 and 0.03 cm (table A.2).

Number of measurements

Using only a single as opposed to the average of three measurements decreased the reliability of all ultrasounds-derived parameters in burn subjects and introduced more significant bias between raters in all subjects (table 4 to A.2). Averaging measurements decreased MDC95 values by approximately 0.1 cm for all QMLT parameters (equivalent to a decrease of 3 to 5% of the mean score depending on the type of compression), and 0.2 cm^2 for RF-CSA (equivalent to 5% of the mean score) in burn subjects (table 3). In healthy subjects, MDC95 values were unchanged regardless of average or single measurements (table A.1).

QMLT compression technique

Using no-compression to measure QMLT generally yielded higher ICC and lower MDC95 values than the maximum-compression technique (table 3 and A.1) in all subjects. In burn subjects, MDC95 values for maximum-compression were on average equivalent to 15% of the mean score, as opposed to an average of 6% of the mean score when using the no-compression technique. Mean differences between raters, although mostly non-significant) were on average higher with maximum-compression than no-compression (0.7 cm vs. 0.2 cm).

QMLT location

Reliability parameters for QMLT were similar between the proximal and distal location and left and right side, irrespective of applied compression technique.

DISCUSSION:

This study investigated the feasibility and reliability of B-mode ultrasound in measuring quadriceps muscle architecture in the acute burn setting with respect to different methodologies. Our main findings show that in the majority of cases quadriceps muscle ultrasound is reliable and feasible to carry out during hospitalisation, and that adapting the methodology to the individual burn patient can improve its feasibility and reliability. Based on these findings, we propose a three-step decision-making tree (figure 4) to guide burn clinicians and researchers in deciding which ultrasound methodology is most appropriate based on different clinical scenarios.

The variability observed in our study can be explained by either subject- or operator-dependent factors. The inclusion of a healthy control group helped identify which of these factors are unique to the burn population. Subject-dependent factors in burn subjects might have been related to pain on compression of open wounds or the impact of oedema, decreasing the image quality and making delineation of structures of interests more difficult. Operator-dependent factors in burn subjects might have been related to the added stress and time pressure, which might have impacted measurement precision to a larger degree than in healthy subjects. However, as this study only produced minor differences in feasibility and reliability between the healthy and burn subjects, it seems more likely that other factors inherent to the methodology might better explain the observed variability.

One such factor, associated with the QMLT protocol, is that measurements were administered on the midline between the superior anterior iliac spice and the superior pole of the patella to facility reproducibility, with operators not allowed to deviate from this midline. However, as there are large inter-individual differences in quadriceps morphology, these measuring points were not always perfectly positioned on top of the rectus femoris muscle, where its thickness is the largest.

Small shifts in transducer position might therefore result in different thickness measurements, although this issue is inherent to all protocols using body landmarks[59].

This study sought to answer several methodological questions. First, we investigated whether it is necessary to repeat each measurement three times and calculate an average, or whether a single measurement would suffice. A single measurement is appealing as it would theoretically shorten the duration of the full measurement procedure. However, our data shows that, while reliability remained high for either method, the averaging of three measurements led to significant reductions in minimal detectable changes in all assessed muscle parameters in burn patients. This is in line with established protocols of quadriceps muscle ultrasound used in different patient populations[43,52–54].

Secondly, we compared two different compression techniques for the measurement of QMLT. While both techniques achieved comparable feasibility, the no-compression technique resulted in superior reliability with smaller minimal detectable changes relative to measured muscle thickness (6% vs. 15% of the mean thickness). The difficulty associated with standardising the amount of tissue compression provides a likely explanation for this observed difference that has been reported previously[43,44,60]. During compression, the position of the rectus femoris in relation to the femur may have been altered, introducing further variability. In the presence of oedema, the maximum-compression technique is nonetheless the more appropriate choice as it is thought to better reflect true muscle size[38,61]. Maximum-compression also offers a solution for exceptionally large thighs that exceed ultrasound penetration. Clinicians using maximum-compression must then bear in mind that changes below 15% are likely due to measurement error, which might potentially be further improved if the same rater repeated measurements in a given patient[43,62]. If that is not feasible, then the use of a curvilinear ultrasound transducer for very large thighs might provide another manner to circumvent the maximum-compression technique. Compared to linear arrays, curvilinear transducers have a deeper penetration at the expense of lower resolution. Despite this trade-off, QMLT and RF-CSA derived through curvilinear transducers have proven equal to those derived through linear transducers, albeit in a different patient population[63,64]. A second alternative in case of too large thighs would be to solely measure the thickness of the more superficial rectus femoris muscle, as done in other trials[43,49,65,66]. However, as the time course of muscle wasting has shown to affect the rectus femoris and intermedius muscle differently over time[65,67], we suggest not to exclude the latter. The fact that the minimal detectable changes during the no-compression manoeuvre in this study all ranged around the 0.2 cm mark regardless of measured thickness, further underlines the importance of using the entire quadriceps layer to be able to detect relative changes earlier. Hence, the alternative use of curvilinear transducers to enable the no-compression technique to determine QMLT in large thighs seems preferred. However, as we have not tested the use of curvilinear transducers in this study, this recommendation should be interpreted with caution.

Thirdly, this study compared two commonly reported locations to measure QMLT, the halfway (proximal) and two third (distal) point of the distance between the superior anterior iliac spine and the upper patellar pole. The reliability data of this study do not support one location over the other, confirming that both points can be reliably used to determine QMLT. In exceptional cases that thigh sizes are too large to allow ultrasound penetration via the no-compression technique, the distal location might prove slightly more feasible as the femur is generally located more superficially than at the proximal site. In all healthy and burn subjects included in this study, we were unable to measure QMLT in eight out of 140 measured thighs at the proximal site, and six out of 140 thighs at the distal site with the no-compression technique. As these are only minor differences in feasibility, one might be tempted to conclude that these two locations are interchangeable. Using multiple locations, including both left and right sides, however, has shown to provide a more complete surrogate measure of whole-body muscle mass[68,69]. Assessing both right and left thighs has additional advantages in the longitudinal assessment of burn subjects. Depending on the post-surgical protocols in place, the thigh, a common graft donor site, might be rendered inaccessible for a certain time due to wound dressings. In our sample, this occurred in three out of 60 measured thighs, and only affected one thigh at a time, leaving the other thigh to be used for the purpose of monitoring of muscle wasting. With respect to QMLT measurement location in burn patients, we therefore recommend the use of multiple locations, both proximal and distal on the right and left thigh.

One potential drawback of ultrasound in particularly major burns is that it needs to take place during wound dressing changes (nearly half of all measurements presented in this report). These dressing changes are one of the most difficult and stressful periods for both the burn team and the patient resulting among others from pain, prolonged wound exposure to air, and range of motion exercises. Consequently, adding muscle assessment to the to-do list during dressing changes is challenging, and has traditionally been postponed to a later timepoint once wound healing is completed. Based on the measured duration of the entire ultrasound procedure in this study, we estimate that it should take clinicians no more than twenty minutes to complete the assessment, making this protocol feasible to take place during dressing changes. The importance of multi-disciplinary teamwork in these cases cannot be overstated. Patient education and coordination with nursing staff, intensivists and anaesthesiologists proved integral to the feasibility of the measurements.

Many previous studies have examined the reliability of QMLT and RF-CSA measurements in the critically-ill population, primarily using the no-compression technique, producing high inter-rater reliability coefficients (ICC >0.9) and small inter-rater differences (<0.1 cm) comparable to this study[37,41–43,46,59,70–72]. However, to the best of our knowledge, no study has to date examined ultrasound as a tool to measure muscle parameters in the burn population. This study is, therefore, the first to demonstrate that ultrasound can be reliably adapted to this population just as well as in the critically-ill population. Our results shows that ultrasound assessment of the quadriceps is feasible and reliable in acute burn patients and that the examined protocol can be used irrespective of wound status, fluid status, and body size.

Strengths and Limitations:

A primary strength of this investigation lies in its multi-centre nature, which allowed the recruitment of a heterogenous sample and the use of different ultrasound machines, thereby enhancing the external validity of our findings. Another strength can be found in the inclusion of a healthy control group, which aided in understanding to which degree subject- and operator-dependent factors caused the observed variability in feasibility and reliability.

In addition to the limitations already discussed, several limitations in this investigation can be identified. One such limitation was the fact that skin markings were not erased between raters, as most measurements had to take place during a very limited time window (during dressing changes) and we were unable to erase skin markings during the same session. While this choice might have overestimated reliability by eliminating potential variability associated with landmarking[59], it is the authors' experience from pilot testing that most of the observed procedural variability did not originate from landmarking, but rather from the ultrasound performance (applied tissue compression and tilt of the transducer)[60]. Nevertheless, in selected burn patients with intact thigh surface, the use of permanent skin markings, as is common in non-burned patient populations, might in fact be desirable for examining longitudinal changes[41].

Another shortcoming of this investigation is that the reliability analysis only focussed on measurements within 72 hours of admission, thereby limiting our ability to comment on the reliability of this ultrasound protocol at later stages of recovery.

While this remains a subject of future research, no additional factors are theoretically present that could impact the reliability at later stages of recovery that did not exist during the admission assessment. Conversely, it is during the early days of a burn centre admission that muscle ultrasound assessment is most challenging and clinical priorities are elsewhere. Rather than delaying or entirely skipping the baseline assessment of muscle, we demonstrate that it is reliable during this crucial phase.

Lastly, as is the case with many clinical investigations, this study was based on the measurements of two raters. Including more raters was not feasible due to the short time-window in which measurements had to take place. While this could have limited our ability to extrapolate our findings to the wider population of raters in the clinical setting, other reports of intensive-care workers have previously demonstrated the reliability of quadriceps muscle ultrasound irrespective of the assessor’s level of expertise[45,66,78]. For the same reason of time, we chose to test inter-rater as opposed intra-rater reliability, as this was more applicable to both participating trial sites, where the care of burn patients traditionally involves multiple clinicians of the same burn care team.

Future directions:

This study lays a well-needed foundation for the many remaining clinical questions surrounding muscular ultrasound in the burn population. Future research should address the reliability of ultrasound at later stages of hospitalisation. Whether ultrasound-derived parameters of muscle size at baseline or over time, correlate with clinical outcomes, such as mortality, duration of mechanical ventilation, nutritional status, etc, forms another clinical question that may guide clinicians in deciding when and how regularly to use of ultrasound for muscular assessment. Finally, the two parameters of interest in this study (QMLT and RF-CSA) were both quantitative in nature. But ultrasound can also measure the echogenicity, which, albeit less researched, may provide a better picture of the qualitative aspects of muscle[65,65].

CONCLUSION

Despite its importance as a metabolic reserve, muscle mass has traditionally not been part of the admission assessment in burn care. In this multi-centre study, we demonstrate that ultrasound measures of quadriceps muscle architecture are feasible and reliable at baseline and can be adapted to different clinical scenarios commonly encountered in the burn setting.

SUPPLEMENTARY MATERIAL

Appendix A

DECLARATION OF INTEREST

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TABLES

Inclusion criteria	Exclusion criteria
Age ≥18 years	Electrical burns
%TBSA ≥10% (with the presence of at least deep second-degree burns)	Associated injury (e.g. lower limb fracture) (interfering with ability to exercise)
Burn centre admission ≤72 hours	Central neurological, peripheral neuromuscular disorders
	Psychological disorders interfering with cooperation
	Diabetes Mellitus type 1
	Pregnancy
	Palliative care

Table 1. Eligibility criteria
TBSA, Total body surface area

	Burn subjects (n = 20)	Healthy subjects (n = 20)	t-test
Clinical trial site 1[a] / trial site 2[b]	9 / 11	/	
Females / Males	4 / 16	10/10	
Age (years)[c]	50 [42.5 – 57.4]	42 [36.3 – 48.4]	*p*=0.11
BMI[c]	27 [25 – 29]	28 [25 – 31]	*p*=0.54
TBSA (%)	32 [22.9 – 40.2]	/	
Days postburn	1.5 [1.1 – 2]	/	
Mechanically ventilated (n)	10	/	
Open wounds (%)	47.5 (19 thighs)	/	
Net fluid balance (ml)	+2410 [1058 – 3762]	/	

Table 2. Characteristics of tested subjects.
Data displayed as mean [95%CI]. BMI, body mass index; TBSA, total body surface area
[a]Refers to the burn centre of ZNA Stuivenberg, Antwerp; [b]Refers to the burn centre of the Military Hospital Queen Astrid, Brussels; [c]Between groups differences tested by two-sample t-tests.

	No. of measurements	Weeks since admission (mean)	QMLT (no compression)		QMLT (max. compression)		RF-CSA
			Proximal location	Distal location	Proximal location	Distal location	
Admission	20	0	92.5%	97.5%	90.0%	87.5%	100%
All FU	30[a]	5.8	95.0%	95.0%	91.7%	91.7%	95.0%
1st FU	16	3.6	90.6%	90.6%	84.4%	84.4%	90.6%
2nd FU	11	6.8	100%	100%	100%	100%	100%
3rd FU	3	14.0	100%	100%	100%	100%	100%
Discharge	16	6.2	100%	100%	100%	100%	100%
TOTAL	50	/	94.0%	96.0%	91.0%	90.0%	97.0%

Table 3. Percentage of successfully completed ultrasound measurements in burn subjects throughout burn centre stay.
QMLT, quadriceps muscle layer thickness; RF-CSA, rectus femoris cross sectional area; FU, follow-up measurements
[a]Refers to 16 subjects

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The most important data is already mentioned in the text…

		ICC [95%CI]		MDC95			
		Average[a]	Single[b]	Average[a]	%mean	Single[b]	%mean
RIGHT	**QMLT (no compression, proximal location)**	0.994 [0.986-0.998]	0.988 [0.97-0.995]	0.22 cm	5.4	0.31 cm	7.8
	QMLT (max. compression, proximal location)	0.98 [0.945-0.993]	0.957 [0.885-0.984]	0.29 cm	16.6	0.43 cm	24.2
	QMLT (no compression, distal location)	0.991 [0.976-0.996]	0.98 [0.95-0.992]	0.23 cm	7.6	0.34 cm	11.4
	QMLT (max. compression, distal location)	0.985 [0.961-0.995]	0.975 [0.934-0.991]	0.18 cm	14.2	0.25 cm	18.8
	RF-CSA	0.99 [0.974-0.996]	0.973 [0.934-0.989]	0.22 cm^2	7.3	0.35 cm^2	11.8
LEFT	**QMLT (no compression, proximal location)**	0.99 [0.973-0.996]	0.978 [0.945-0.992]	0.29 cm	7.5	0.42 cm	11
	QMLT (max. compression, proximal location)	0.971 [0.901-0.99]	0.953 [0.758-0.986]	0.31 cm	18.4	0.40 cm	23.2
	QMLT (no compression, distal location)	0.995 [0.988-0.998]	0.986 [0.965-0.994]	0.17 cm	5.6	0.28 cm	9.4
	QMLT (max. compression, distal location)	0.99 [0.974-0.996]	0.977 [0.843-0.994]	0.15 cm	10.7	0.22 cm	16
	RF-CSA$_{ln}$[c]	0.955 [0.888-0.982]	0.895 [0.756-0.957]	0.37 cm^2	12.7	0.57 cm^2	19.7

Table 3. Intraclass correlations coefficients and minimal detectable changes in burn subjects.
QMLT, quadriceps muscle layer thickness; RF-CSA, rectus femoris cross sectional area
[a]Refers to the average of three measurements; [b]Refers to the first of three measurements; [c]ICC values were backtransformed to derive MDC95 of RF-CSA of the left thigh.

		Systematic bias[a]				Proportional bias			
		Average[b]		Single[c]		Average[b]		Single[c]	
		Mean Δ	*p*-Value	Mean Δ	*p*-Value	β Co-eff	*p*-Value	β Co-eff	*p*-Value
RI	**QMLT (no compression, proximal location)**	0.02 cm	0.665	0.03 cm	0.357	-0.031	0.118	0.019	0.614
	QMLT (max. compression, proximal location)	-0.08 cm	0.124	-0.08 cm	0.111	0.029	0.674	0.027	0.704

	QMLT (no compression, distal location)	0.02 cm	0.683	0.00 cm	0.958	0.084	0.066	0.075	0.125
	QMLT (max. compression, distal location)	-0.03 cm	0.309	-0.01 cm	0.645	0.001	0.987	0.025	0.671
	RF-CSA	-0.04 cm^2	0.287	-0.04 cm^2	0.286	0.058	0.221	0.115	0.027*
LEFT	**QMLT (no compression, proximal location)**	<0.01 cm	0.999	-0.02 cm	0.729	0.091	0.062	0.098	0.049*
	QMLT (max. compression, proximal location)	-0.11 cm	0.022*	-0.13 cm	0.003*	0.067	0.373	0.044	0.469
	QMLT (no compression, distal location)	-0.01 cm	0.726	-0.01 cm	0.803	-0.052	0.106	-0.072	0.063
	QMLT (max. compression, distal location)	-0.04 cm	0.131	-0.08 cm	0.001*	-0.019	0.685	-0.038	0.343
	RF-CSA$_{ln}$	-0.01 cm^2	0.598	-0.02 cm^2	0.459	0.036	0.725	0.009	0.935

Table 4. Systematic and proportional bias in burn subjects.

QMLT, quadriceps muscle layer thickness; RF-CSA, rectus femoris cross sectional area

[a] Systematic bias refers to the mean differences between raters, subtracting rater 1 - rater 2; [a] Refers to the average of three measurements; [b] Refers to the first of three measurements; *p value < 0.05

FIGURES

See separate files.