ANESTHESIOLOGY

Changes in Respiratory Muscle Thickness during Mechanical Ventilation

Focus on Expiratory Muscles

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ABSTRACT

Background: The lateral abdominal wall muscles are recruited with active expiration, as may occur with high breathing effort, inspiratory muscle weakness, or pulmonary hyperinflation. The effects of critical illness and mechanical ventilation on these muscles are unknown. This study aimed to assess the reproducibility of expiratory muscle (*i.e.*, lateral abdominal wall muscles and rectus abdominis muscle) ultrasound and the impact of tidal volume on expiratory muscle thickness, to evaluate changes in expiratory muscle thickness during mechanical ventilation, and to compare this to changes in diaphragm thickness.

Methods: Two raters assessed the interrater and intrarater reproducibility of expiratory muscle ultrasound (n = 30) and the effect of delivered tidal volume on expiratory muscle thickness (n = 10). Changes in the thickness of the expiratory muscles and the diaphragm were assessed in 77 patients with at least two serial ultrasound measurements in the first week of mechanical ventilation.

Results: The reproducibility of the measurements was excellent (interrater intraclass correlation coefficient: 0.994 [95% CI, 0.987 to 0.997]; intrarater intraclass correlation coefficient: 0.992 [95% CI, 0.957 to 0.998]). Expiratory muscle thickness decreased by 3.0 ± 1.7% (mean ± SD) with tidal volumes of 481 ± 64 ml ($P < 0.001$). The thickness of the expiratory muscles remained stable in 51 of 77 (66%), decreased in 17 of 77 (22%), and increased in 9 of 77 (12%) patients. Reduced thickness resulted from loss of muscular tissue, whereas increased thickness mainly resulted from increased interparietal fasciae thickness. Changes in thickness of the expiratory muscles were not associated with changes in the thickness of the diaphragm ($R^2 = 0.013$; $P = 0.332$).

Conclusions: Thickness measurement of the expiratory muscles by ultrasound has excellent reproducibility. Changes in the thickness of the expiratory muscles occurred in 34% of patients and were unrelated to changes in diaphragm thickness. Increased expiratory muscle thickness resulted from increased thickness of the fasciae.

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EDITOR'S PERSPECTIVE

What We Already Know about This Topic

- The respiratory muscle pump consists of three primary groups controlling ventilation: the diaphragm, the primary muscle of inspiration; the accessory inspiratory (parasternal, external intercostal, scalene, and sternocleidomastoids); and the expiratory muscles (lateral abdominal wall muscles, internal intercostals, and the transverse thoracic).
- Mechanical ventilation for respiratory failure has documented adverse effects on the diaphragm and other inspiratory muscles, but its impact on expiratory muscles has not been well characterized.
- Focusing on ultrasound-derived measurement of the thickness of the lateral abdominal wall musculature (transversus abdominus, internal, and external oblique muscles) in mechanically ventilated patients in a mixed medical surgical intensive care unit, the authors performed a reproducibility study investigating inter- and intrarater variability (30 patients) and correlation with tidal volume (10 patients) on muscle thickness. They then performed a cohort study (77 patients with at least two serial measurements in the first week of ventilation) to investigate the temporal evolution of such changes, grouping patients into those that decreased, had no change, or increased over time using threshold values from the reproducibility cohort.

What This Article Tells Us That Is New

- Inter- and intrarater reproducibility was strong (intraclass correlation coefficients 0.994 [95% CI, 0.987 to 0.997] and 0.992 [95% CI, 0.957 to 0.998], respectively).
- Muscle thickness increased by 3.2% after increasing lung volume by a mean ± SD of 481 ± 64 ml.
- Although muscle thickness remained stable in the majority of subjects, it decreased in 22% and increased in 12% with no association with changes in diaphragmatic thickness. Exploratory analyses suggest no relation with a variety of clinical or physiologic parameters or medications.
- Time-dependent decreases in thickness resulted from muscle loss, whereas increases largely resulted from increases in thickness of the interparietal fasciae.

This article is featured in "This Month in Anesthesiology," page 1A. This article is accompanied by an editorial on p. 680. Supplemental Digital Content is available for this article. Direct URL citations appear in the printed text and are available in both the HTML and PDF versions of this article. Links to the digital files are provided in the HTML text of this article on the Journal's Web site (www.anesthesiology.org). Z.-H.S. and H.d.V. contributed equally to this article.

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The respiratory muscle pump is a vital organ that drives alveolar ventilation. The respiratory muscle pump consists of several muscle groups: the diaphragm, which is the main muscle for inspiration; the accessory inspiratory muscles, including the parasternal, external intercostal, scalene, and sternocleidomastoid muscles; and the expiratory muscles, including the lateral abdominal wall muscles, the internal intercostal muscles, and transverse thoracic muscle.[1–6] With impending respiratory failure, mechanical ventilation is a life-saving intervention to support the respiratory pump. However, it is now recognized that mechanical ventilation may have adverse effects on the respiratory muscles.[7–17] Ultrasound studies revealed time-dependent loss of diaphragm thickness.[15,17] Moreover, studies analyzing diaphragm biopsies of ventilated intensive care unit (ICU) patients revealed diaphragm fiber atrophy, impaired contractile protein function, injury, and inflammation.[10–14]

The effects of mechanical ventilation on the expiratory muscles are largely unknown.[1] This is surprising, because these muscles play an important role in airway clearance, prevention of atelectasis,[18–20] and maintaining alveolar ventilation with high breathing effort.[1,6,21] Weakness of the expiratory muscles has been associated with reintubation, rehospitalization, and mortality.[22–25] Although both the diaphragm and the lateral abdominal wall muscles are important components of the respiratory muscle pump, it is unknown whether these muscles respond in a similar fashion to mechanical ventilation and critical illness, *i.e.*, if atrophy of the diaphragm correlates with atrophy of the lateral abdominal wall muscles.

Ultrasound has become a popular tool to investigate diaphragm thickness in ICU patients.[15,26–28] However, very few studies have applied ultrasound to evaluate the expiratory muscle thickness, and therefore the current study aimed to evaluate feasibility and reproducibility of expiratory muscle thickness measurements in critically ill ventilated patients. Because higher lung volume results in caudal displacement of the diaphragm and may as such expand the abdominal wall, we also aimed to assess the influence of lung volume on expiratory muscle thickness (reproducibility study). Second, we aimed to investigate time-dependent changes in the expiratory muscle thickness in critically ill patients during the first week of mechanical ventilation and to evaluate whether changes in expiratory muscle thickness are associated with changes in diaphragm thickness (cohort study). As an exploratory analysis, we assessed whether patient characteristics were associated with the evolution of expiratory muscle thickness and whether the evolution of expiratory muscle thickness was associated with clinical outcomes.

Materials and Methods

Study Design and Patients

This prospective, observational study was conducted from February 2017 to October 2020 in a mixed medical surgical academic ICU (Amsterdam University Medical Centers, Location VUmc, Amsterdam, The Netherlands). An opt-out approach for the consent from subjects or their legal representative was used because mechanical ventilation is a requirement for eligibility, and ultrasound measurement is daily routine in this medical center. The study protocol was approved by the local institutional ethics committees and registered on ClinicalTrials.gov (NCT04333186, April 2020). The study was performed in accordance with the ethical standards set forth in the 2008 Declaration of Helsinki and its later amendments. In the current study, the lateral abdominal wall muscles (transversus abdominis muscle, internal oblique muscle, and external oblique muscle) are referred to as the expiratory muscles, unless otherwise stated.

This study consisted of two substudies (fig. 1). The reproducibility study tested the feasibility and reproducibility of expiratory muscle ultrasound and assessed the effects of lung volume on expiratory muscle thickness. The cohort study aimed to investigate the evolution of expiratory muscle thickness during the mechanical ventilation in critically ill patients. We used the data from the reproducibility study to determine the threshold values to categorize patients into three groups of muscle thickness changes (decreased, no change, and increased) in the cohort study.

In the reproducibility study, adult (more than 18 yr old) ventilated critically ill patients were recruited. Exclusion criteria were past medical history of neuromuscular disorders, abdominal wounds at the proposed location of the ultrasound probe, and pregnancy. Two raters (M.H. and Z.-H.S.) performed the measurements; both had extensive experience in respiratory muscle ultrasound. In one single session, each rater performed ultrasound measurements at the same anatomical location as marked by the first rater within a ±5-min interval. Images were stored for later offline measurements. An intrarater reliability test was performed in a subset of eight patients, in which each rater repeated measurements at the previously marked anatomical site 5 min after the interrater session. The raters were blinded to their own and each other's measurements.

To assess the influence of lung volume on expiratory muscle thickness, ultrasound measurements were repeated in a subgroup of 10 patients in the reproducibility study that showed no signs of respiratory muscle activity during an end-expiratory breath hold. Expiratory muscle thickness was measured at end-inspiratory lung volume (during the last second of a 3-s end-inspiratory occlusion) and at end-expiratory lung volume (during the last second of 3-s end-expiratory occlusion). The difference in lung volume (*i.e.*, delivered tidal volume) between these two measurements was recorded from the ventilator (Servo-U, Sweden). These measurements were performed by a single rater (Z.-H.S.).

The cohort study aimed to assess time-dependent changes of expiratory muscle thickness in critically ill

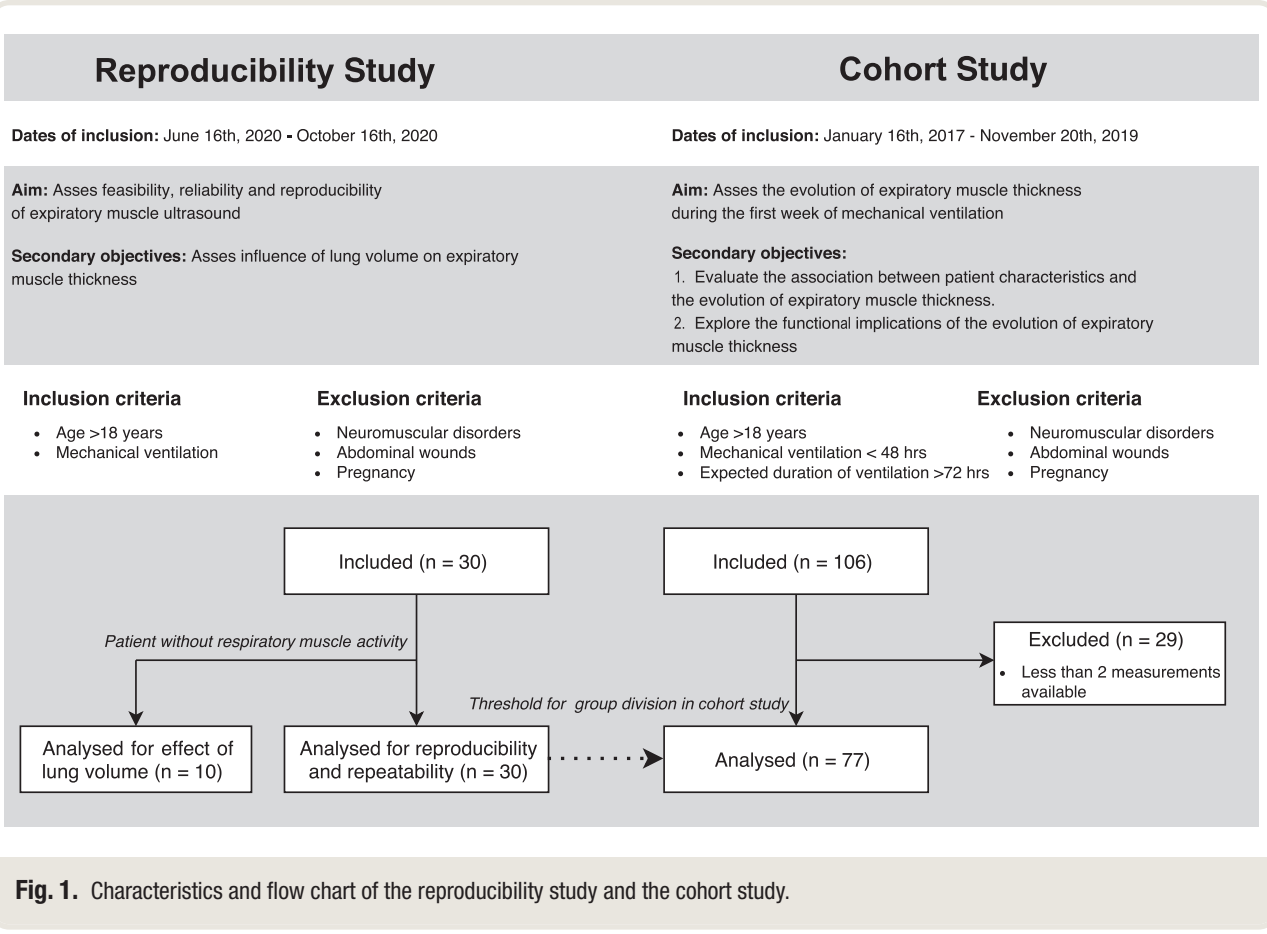

Fig. 1. Characteristics and flow chart of the reproducibility study and the cohort study.

mechanically ventilated patients. Patients admitted to the ICU were screened daily between 9 and 10 AM (Monday through Thursday). Patients were eligible for enrolment within 48 h after intubation if the treating physician expected them to remain ventilated for more than 72 h. Exclusion criteria were identical to the reproducibility study. The medical team followed standard clinical protocols for mechanical ventilation and weaning.

Ultrasound Examination

All measurements were performed on the right side with the patient in the supine position. The probe position was marked on the skin after the first measurements to ascertain an identical anatomical location in subsequent measurements. B-mode ultrasound images were acquired using a high frequency (10 to 15 MHz) linear array transducer (CX50, Philips, USA). Expiratory muscle thickness was measured at end inspiration (the frame with minimum thickness within one breath) and included the three individual muscle layers (fig. 2). The mean of the three breaths in one assessment session was saved for further analysis. The measurements were performed daily from Monday to Friday until patients were extubated, discharged from the ICU, transferred to another hospital, or deceased, whichever came first. Detailed methods for ultrasound measurements are described in the legend of figure 2.

Clinical Factors

Patient characteristics for the cohort study were collected at the time of inclusion. Mechanical ventilation–related variables including mode of mechanical ventilation, positive end-expiratory pressure (PEEP), inspiratory tidal volume, applied driving pressure (peak pressure–PEEP), drug administration, and fluid balance were extracted from the electronic patient record on an hourly basis from the moment of recruitment to the end of the study period. Patients were followed up until hospital discharge.

Statistical Analysis

Normality of the distribution of the studied variables was assessed visually on normal probability plots. The data are expressed as mean ± SD, median [interquartile range], or frequency (percentage), as appropriate. We used a two-sided significance level of 5% for all analyses. Mean imputation was used for clinical characteristics that were missing. Missing data on muscle thickness during the study period were considered missing at random and were not imputed.

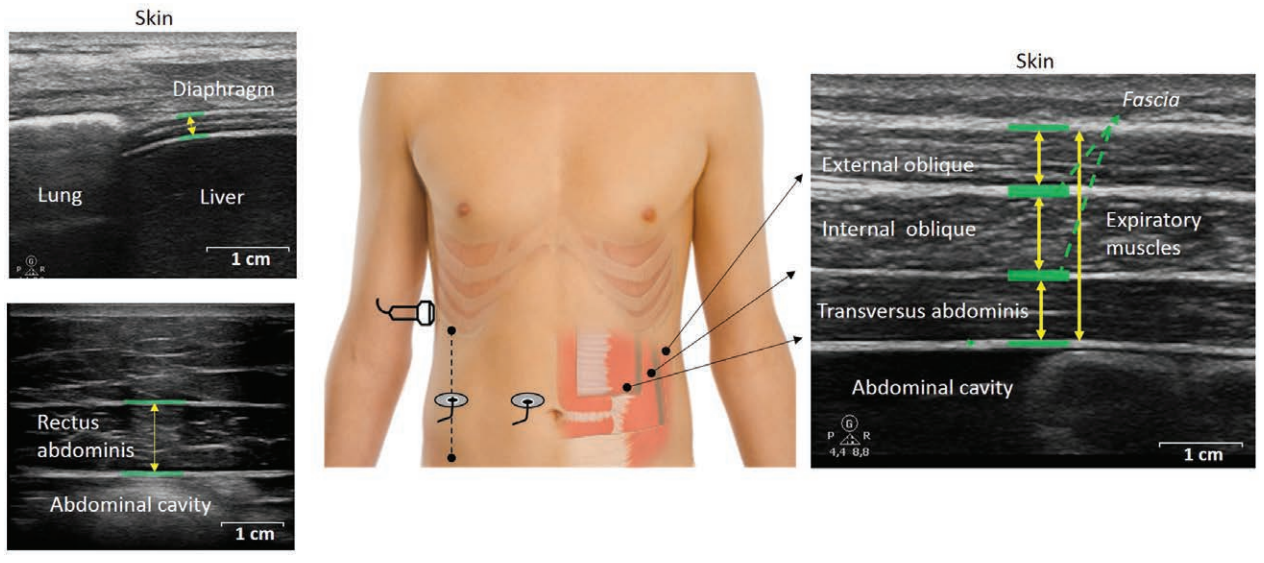

Fig. 2. (*Top left*) Representative ultrasound image of the diaphragm. The muscle thickness was measured as the distance between the inner layers of the fasciae at end expiration. The probe was located at the zone of apposition between the midaxillary or anterior–axillary line in the 8th to 11th intercostal space. (*Bottom left*) Representative ultrasound image of the rectus abdominis muscle. The measurement of the muscle thickness was performed at end inspiration and was performed perpendicular to the inner layer of the muscle fascia. The probe was located at approximately 2 to 3 cm above the umbilicus and 2 to 3 cm lateral from the midline. (*Right*) Representative ultrasound image of the lateral abdominal expiratory muscles, showing three muscles from the top to bottom: external oblique, internal oblique, and transversus abdominis muscles. The measurements of the thickness of expiratory muscles and individual layers were performed at end inspiration and were performed perpendicular to the inner side of the muscle fascia (*yellow arrow*). The muscle fasciae are presented as *green lines*. The probe for the lateral abdominal muscles measurement was located approximately on the anterior axillary line, midway between the inferior border of the rib cage and the iliac crest.

For the reproducibility study, intraclass correlation coefficient models with measures of consistency were constructed (two-way random for interrater and one-way random for intrarater, respectively). The intraclass correlation coefficient values of the average measures were reported as intraclass correlation coefficient (95% CI). Repeatability coefficient, bias, and the upper/lower limit of agreement were calculated between raters and within each rater.[29–31] We calculated the required sample size for the reproducibility study assuming an intraclass correlation coefficient between two raters of at least 0.85 with 95% CI of width of at most 0.2 and intrarater intraclass correlation coefficient of 0.95 with 95% CI width of at most 0.1. Under these assumptions, 30 patients were required to evaluate interrater reliability, and 16 patients were required to evaluate intrarater reliability (eight patients for each rater to measure twice).

For the cohort study, we used the limits of agreement from the reproducibility study to obtain thresholds for changes in expiratory muscle thickness that were unlikely to arise from measurement variance alone (*i.e.*, to determine the minimal difference in muscle thickness that is likely to be attributable to biologic processes such as atrophy and hypertrophy, as opposed to measurement variance). We used linear regression to estimate the average change in the expiratory muscle thickness for each patient over the first week of study. The study population was divided into three subgroups based on the estimated change in thickness on the last day of measurement (Supplemental Digital Content 1, http://links.lww.com/ALN/C566, fig. E1). If the thickness of the expiratory muscles increased by at least 15% compared with its own baseline on the last day of measurement, a patient was assigned to the *increase* group. If thickness of the expiratory muscle decreased by at least 15%, a patient was assigned to the *decrease* group. If relative change was less than 15% in either direction, a patient was assigned to the *no change* group.

To compare the thickness of the expiratory muscles at different days between the three groups, a linear mixed model design was used with a fixed effect for day of study, group, and group-by-day interaction, a random effect for each patient, and baseline thickness as a covariate. A Bonferroni *post hoc* correction was applied for the pairwise comparisons. Differences in patient characteristics, baseline ventilator parameters, and clinical outcomes between these three groups were analyzed using ANOVA with *post hoc* Tukey honest significant difference test, the Kruskal–Wallis test with Dunn *post hoc* test, or chi-square tests with *post*

hoc tests as appropriate. *Post hoc* tests were only performed when the overall test was significant.

The association between changes in expiratory muscle thickness and diaphragm thickness was analyzed with a linear regression model, using the estimated difference in thickness on the last day of measurement in the first week for both the diaphragm and the expiratory muscles (Supplemental Digital Content 1, http://links.lww.com/ALN/C566, fig. E1). Given the exploratory nature of the cohort study, no formal sample size calculation was performed. We planned to enroll 100 subjects.

Sensitivity Analyses

Sensitivity analysis models were added after suggestions by the reviewers to evaluate whether the different thresholds to define the three groups derived from the reproducibility study would have resulted in different conclusions. Because patients could have had a variable number of measurements within the first week, we additionally used linear mixed models to estimate the average change of expiratory muscle thickness over time. Linear mixed models were also added after suggestions from the reviewers to test whether changes in thickness were related to baseline characteristics. Linear regression was used to test for associations between the patients' average changes in expiratory muscle thickness (as a continuous parameter) over the first week and clinical outcomes. This approach has more power and does not depend on thresholds. The sensitivity analyses are presented in more detail in Supplemental Digital Content 1 (http://links.lww.com/ALN/C566). The data were analyzed with R version 4.0.2 (R Foundation for Statistical Computing, Austria) and SPSS version 26 (SPSS for Windows, IBM Corp., USA).

Results

Reproducibility Study: Reproducibility of Expiratory Muscle Thickness Measurements in Ventilated Subjects and Effect of Lung Volume

Thirty critically ill patients (25 male, 62 ± 17 yr, body mass index = 25.3 ± 3.2 kg/m^2) were enrolled, and expiratory muscle thickness measurements were obtained in all of these patients. The repeatability and reproducibility for the measurements between and within raters are reported in table 1. Intraclass correlation coefficients for the ultrasound measurements of the expiratory muscles were excellent for both interrater and intrarater (interrater intraclass correlation coefficient: 0.994 [95% CI, 0.987 to 0.997], intrarater intraclass correlation coefficient for two raters: 0.998 [95% CI, 0.992 to 1.000] and 0.992 [95% CI, 0.957 to 0.998], respectively). The means of difference caused by repeated measurements were 3.1% with limits of agreement from −13.1 to 6.8% between two raters and 1.4% with limits of agreement from −9.4 to 12.1% for the same rater (table 1).

To assess the effect of lung volume on the expiratory muscle thickness, a subgroup of 10 patients (57 ± 20 yr, all male, body mass index = 23.6 ± 2.6 kg/m^2) was analyzed. The mean tidal volume change between two measurements within a subject was 481 ± 64 ml. Mean of difference between the expiratory muscle thickness measured at high lung volume and low lung volume was −0.5 ± 0.4 mm (16.1 ± 2.9 mm *vs.* 16.7 ± 3.2 mm, $P < 0.001$). The thickness decreased by 3.2% after increasing lung volume, with limits of agreement between 0.1 and 6.5% (table 1).

Cohort Study: Time-dependent Changes of Expiratory Muscle Thickness in Critically Ill Mechanically Ventilated Patients

During the study period, 106 patients were enrolled, of whom 29 patients were extubated or died before the second ultrasound measurement was obtained. Accordingly, 77 patients with 331 ultrasound measurements were analyzed. The first ultrasound measurement was performed at a median of 21 [15 to 28] hours after the start of mechanical ventilation. The demographic and clinical characteristics of the study population are presented in table 2.

Baseline Thickness of the Expiratory Muscles

The baseline median thickness of the expiratory muscles was 13.1 [10.2 to 16.1] mm. We observed a statistically significant association between age and baseline thickness of the expiratory muscles (0.16 mm decrease per year, 95% CI, 0.13 to 0.19; $R^2 = 0.251$; $P < 0.001$) but not between body

Table 1. Reproducibility of the Expiratory Muscle Thickness Measurements in Mechanically Ventilated Patients

	Subjects, n	Mean ± SD, mm	Repeatability Coefficient, mm	Bias ± SD, %	95% Limits of Agreement, %	Intraclass Correlation Coefficient
Interrater	30	13.1 ± 4.7	1.6	−3.1 ± 5.1	−13.1 to 6.8	0.994 (0.987 to 0.997)
Intrarater (M.H.)	8	13.0 ± 6.0	1.0	1.4 ± 5.5	−9.4 to 12.1	0.998 (0.992 to 1.000)
Intrarater (Z.-H.S.)	8	11.7 ± 4.5	1.6	1.2 ± 6.5	−11.4 to 13.8	0.991 (0.957 to 0.998)
Lung volume (high *vs.* low)	10	16.4 ± 3.0	1.2	−3.2 ± 1.7	−6.5 to 0.10	0.992 (0.969 to 0.998)

The data for bias and 95% limits of agreement were generated from Bland–Altman plots, which plot the difference between the two measurements as a percentage. The bias is presented as the mean ± SD, and the 95% limits of agreement are presented as lower limit of agreement–upper limit of agreement. Intraclass correlation coefficient is presented with 95% CI.

Table 2. Clinical Characteristics of the Study Patients and Subgroups According to the Thickness Changes in the Expiratory Muscles

		Change in Muscle Thickness of the Expiratory Muscles			
Variables	**Overall (n = 77)**	**No Change (n = 51)**	**Decrease (n = 17)**	**Increase (n = 9)**	***P* Value**
Age, yr	66 [56–70]	64 [56–70]	64 [54–68]	71 [51–73]	0.526
Sex, male	57 (74)	40 (78)	10 (59)	7 (78)	0.269
Body mass index, kg/m^2	25.0 [23.0–28.8]	24.5 [22.5–29.2]	26.4 [23.9–27.7]	25.2 [23.9–29.8]	0.742
C-reactive protein, mg/l	153 [98–243]	149 [97–222]	168 [97–310]	172 [107–243]	0.847
APACHE II score	21 [13–22]	21 [13–23]	22 [12–22]	18 [11–23]	0.906
Sepsis at admission, n (%)	21 (27)	14 (28)	4 (24)	3 (33)	0.866
Septic shock at admission, n (%)	17 (22)	12 (24)	2 (12)	3 (33)	0.411
Indications for mechanical ventilation, n (%)					0.445
Respiratory dysfunction	25 (32)	14 (27)	7 (41)	4 (44)	
Cardiovascular dysfunction	24 (31)	19 (37)	3 (18)	2 (22)	
Neurologic dysfunction	12 (16)	10 (20)	2 (12)	0 (0)	
Other organ dysfunction	16 (21)	8 (16)	5 (29)	3 (34)	
Medical history, n (%)					
COPD	7 (9)	4 (8)	2 (12)	1 (11)	0.866
Hypertension	20 (26)	13 (25)	5 (29)	2 (22)	0.915
Diabetes	18 (23)	15 (29)	1 (6)	2 (22)	0.139
Ventilator settings at baseline					
Controlled mode of ventilation, h	15 [8–22]	15 [8–21]	16 [6–28]	13 [5–25]	0.901
PEEP, cm H_2O	8 [6–12]	8 [6–12]	8 [5–11]	10 [5–15]	0.576
Total respiratory rate, breaths/min	22 [19–25]	23 [29–26]	20 [18–24]	22 [18–24]	0.191
Driving pressure, cm H_2O	13 [10–16]	14 [11–16]	12 [9–23]	13 [8–15]	0.654
Tidal volume per kg of ideal body weight, ml/kg ideal body weight	6.5 [6.1–7.5]	6.4 [6.1–7.1]	7.4 [6.1–8.2]	6.6 [5.9–7]	0.342
P_{AO_2}/F_{IO_2} ratio	217 [156–317]	235 [152–342]	199 [167–321]	208 [142–234]	0.383
Ventilator settings over the first week*					
Controlled mode of ventilation, h	41 [26–68]	41 [26–55]	48 [26–102]	33 [24.5–54]	0.316
PEEP, cm H_2O	8 [6–11]	8 [6–11]	8 [5–12]	10 [7–10]	0.683
Total respiratory rate, breaths/min	22 [20–25]	24 [18–26]	22 [18–24]	20 [16–23]	0.079
Driving pressure, cm H_2O†	12 [9–15]	12 [9–15]	10 [8–18]	10.[6–12]	0.129
Tidal volume, ml/kg ideal body weight	6.5 [6.1–7.6]	6.5 [6.1–7.8]	7.4 [6.1–7.8]	6.5 [6.1–8.3]	0.320
P_{AO_2}/F_{IO_2} ratio	212 [169–289]	228 [173–310]	198 [166–229]	189[155–244]	0.225
Medical treatment over the first week					
Neuromuscular blockers, n (%)	30 (39)	20 (39)	6 (35)	4 (44)	0.900
Corticosteroids, n (%)	24 (31)	15 (29)	6 (35)	3 (33.3)	0.892
Opioids, n (%)	69 (90)	44 (86)	16 (94)	9 (100)	0.363
Vassopressors, n (%)	74 (96)	49 (96)	16 (94)	9 (100)	0.762
Sedatives, n (%)	74 (96)	49 (96)	16 (94)	9 (100)	0.762
Fluid balance, ml	1,935 [–19–4,961]	1,935 [24–5,545]	2,355 [–1,202–4,961]	1,846 [487–4,062]	0.814

Data are expressed as median [interquartile range] or n (%). Missing data were in the range of 2.6 to 6.5%: C-reactive protein (3/74), body mass index (4/73), and tidal volume of ideal body weight (2/75), respectively.

*Ventilator settings over the first week were collected on an hourly basis, and the median of each patients was used for analysis. The coefficients varied from subject to subject, but mostly ranged from 15 to 25%. †The driving pressure applied by the ventilator was defined as peak pressure minus positive-end expiratory pressure (PEEP), both for controlled and partially assisted modes of ventilation.

APACHE II, Acute Physiology and Chronic Health Evaluation II; COPD, chronic obstructive pulmonary disease; F_{IO_2}, fractional inspired oxygen tension; PEEP, positive end-expiratory pressure.

mass index and baseline thickness and Acute Physiology and Chronic Health Evaluation II and baseline thickness. There was no statistically significant difference in expiratory muscle thickness between male and female patients (13.4 [10.2 to 17.7] mm *vs.* 12.2 [9.7 to 15.1] mm, respectively; $P = 0.293$).

Changes in Expiratory Muscle Thickness with Mechanical Ventilation

The limits of agreement of the Bland–Altman plot, constructed on relative differences obtained from the reproducibility study (Supplemental Digital Content 1, http://links.lww.com/ALN/C566, fig. E3), suggested that within-patient variation due to measurement variance would fall below the 15% limit (Supplemental Digital Content 1, http://links.lww.com/ALN/C566, tables E1 and E2). We reasoned that differences above this threshold were likely to be attributed to biologic processes such as atrophy or hypertrophy.

Over the first week of mechanical ventilation, the thickness of the expiratory muscles remained stable in 51 (66%) patients, decreased in 17 (22%) patients, and increased in 9 (12%) patients (fig. 3, Supplemental Digital Content 2 [http://links.lww.com/ALN/C567], 3 [http://links.lww.com/

ALN/C568], and 4 [http://links.lww.com/ALN/C569], which are representative videos of the thickness changes over time from one patient in each group). Significant changes in the expiratory muscle thickness developed as early as the second day of the study. At the second day of the study, the thickness of the expiratory muscles decreased from 15.7 ± 4.3 to 13.9 ± 4.2 mm in the decrease group ($P = 0.013$) and increased from 13.7 ± 7.4 to 18.1 ± 10.5 mm in the increase group ($P = 0.007$; Supplemental Digital Content 1, http://links.lww.com/ALN/C566, table E3). No significant differences in clinical parameters, physiologic parameters, or medication was found among the three groups (table 2). In addition, thickness at baseline was not associated with changes in muscle thickness during the first week ($P = 0.891$).

Differences between Changes in Muscular and Fascia Tissue of the Expiratory Muscles

Thickness measurements of the expiratory muscles include the superficial and deeper interparietal fasciae (fig. 2). Changes in muscle thickness could thus (partly) result from changes in the thickness of these fasciae. Changes in the muscular parts and the fascial parts of the expiratory muscles are shown in figure 4. The time-dependent decrease in expiratory muscle thickness resulted from a decrease in muscular tissue; in these patients, thickness of the fascia did not change over time. In contrast, the increase in expiratory muscle thickness largely resulted from an increase in thickness of the two interparietal fasciae.

Correlation between Thickness Changes of the Expiratory Muscles and the Rectus Abdominis Muscle

The rectus abdominis muscle is part of the abdominal wall but has a limited role in active expiration.[1,4] Changes of the rectus abdominis muscle thickness were significantly associated with changes in expiratory muscle thickness, although the correlation was weak ($R^2 = 0.159$; $P < 0.001$; Supplemental Digital Content 1, http://links.lww.com/ALN/C566, fig. E4). Significant associations between changes in total expiratory muscles thickness and their individual layers were detected ($P < 0.001$), with moderate to

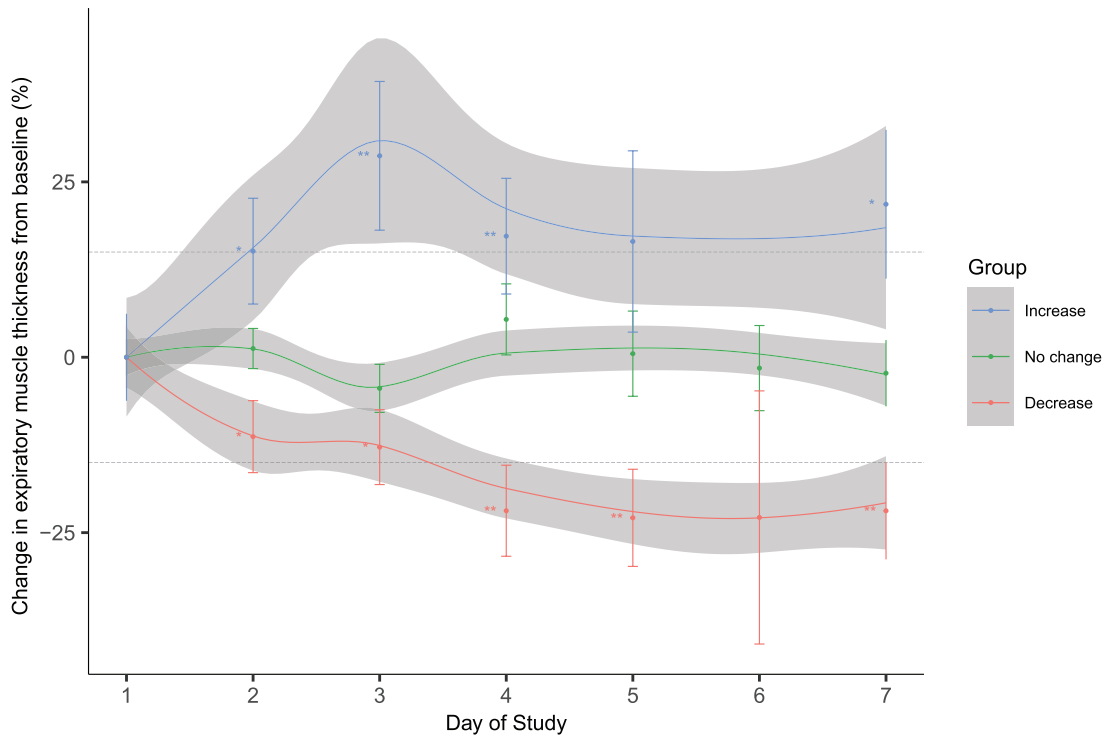

Fig. 3. Expiratory muscle thickness over the first week of mechanical ventilation. Subjects were divided into three groups based on increase or decrease of the estimated thickness of the last measurement within the first week *versus* initial thickness. The estimated thickness was obtained with linear regression using all available measurements for each subject. Therefore, the groups reflect the global changes of each patient. The muscle thickness remained stable (± 15% changes) in 51 (66%) patients, decreased more than 15% in 17 (22%) patients, and increased more than 15% in 9 (12%) patients. Estimated mean and 95% CI are plotted for each group for each day. Trend lines were obtained with local weighted regression. At day 7 of the study, there were 3 patients in the increase group, 8 patients in the decrease group, and 15 patients left in the stable group. At day 6, one measurement was obtained in the decrease group, and no measurements were obtained in the increase group. Compared with the baseline, *$P < 0.05$, **$P < 0.001$.

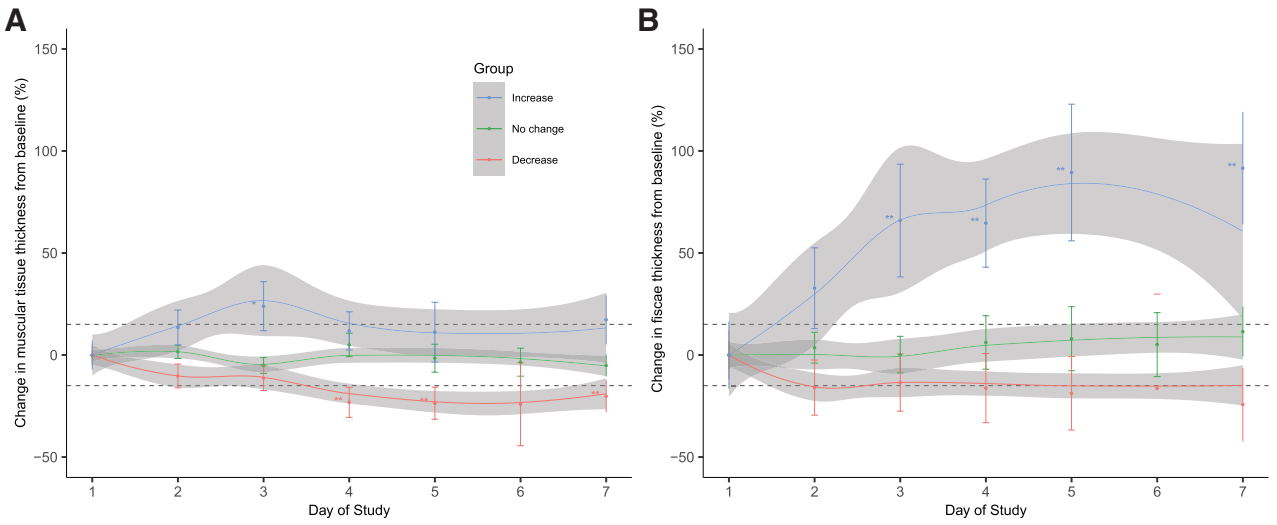

Fig. 4. Thickness of the muscular tissue and the fasciae over the first week of mechanical ventilation. Patients are categorized into three subgroups according to the change in total thickness of the expiratory muscles over time. (*A*) Compared with the baseline thickness, the thickness of the muscular tissue decreased significantly at day 4 and continuously decreased over the following days. The thickness of muscular tissue remained stable in the increase and no change groups. (*B*) Compared with the baseline thickness, the thickness of the fasciae increased significantly on day 3 and continuously increased over the following days. Estimated mean and 95% CI are plotted for each group for each day. Trend lines were obtained with local weighted regression. Compared with the baseline, $*P < 0.05$, $**P < 0.001$.

weak correlation of determination (internal oblique muscle > transversus abdominis muscle > external oblique muscle, with R^2 equal to 0.579, 0.487, and 0.440, respectively).

Correlation between Changes in Thickness of the Expiratory Muscles and Diaphragm

Time-dependent changes in the thickness of the diaphragm were not significantly correlated with the changes in the thickness of the expiratory muscles ($R^2 = 0.013$; $P = 0.332$; Supplemental Digital Content 1, http://links.lww.com/ALN/C566, fig. E5).

Clinical Outcomes

No significant differences in clinical outcomes were found among the three groups defined by time-dependent changes in expiratory muscle thickness (table 3). A sensitivity analysis was performed to assess the associations between slope of change in expiratory muscle thickness (as a continuous variable) and clinical outcomes (Supplemental Digital Content 1, http://links.lww.com/ALN/C566, tables E4 and E5). A significant association was found between the slope of expiratory muscle thickness and hospital length of stay: more negative slopes in the expiratory muscle thickness (*i.e.*, more loss of muscle mass) were associated with increased hospital length of stay. Hospital length of stay increased by 7.4% (95% CI, 1.6 to 13.1%; $P = 0.014$) *per* 0.1 mm/day loss of expiratory muscle mass.

Discussion

This study provides a comprehensive insight into the effects of critical illness and mechanical ventilation on changes in thickness of the most prominent muscle groups of the respiratory pump. These data show that (1) ultrasound is a highly reproducible tool to assess thickness of the expiratory muscles in mechanically ventilated critically ill patients; (2) lung volume in the range of tidal breathing has a significant but small (± 0.5 mm, 3% of the total thickness) effect on expiratory muscle thickness; (3) expiratory muscle thickness decreases in 22%, increases in 12%, and remains stable in 66% of critically ill ventilated patients; (4) the observed increase in thickness of the expiratory muscles mainly results from an increase in thickness of the muscle fasciae; and (5) changes in thickness of the expiratory muscles are not associated with changes in the thickness of the diaphragm. As an explorative endpoint, we observed that loss of expiratory muscle mass during the first week of ventilation was associated with increased hospital length of stay.

Function of the Expiratory Muscles in the ICU

The expiratory muscles are an essential component of the respiratory pump. The lateral abdominal wall muscles are the most prominent expiratory muscles. Activation of the expiratory muscles during breathing occurs when a disbalance develops between load and capacity of the inspiratory muscles, such as with strenuous exercise, low respiratory

Table 3. Clinical Outcomes of the Study Patients and Subgroups According to the Thickness Changes in the Expiratory Muscles

Variables	Overall (n = 77)	No Change (n = 51)	Decrease (n = 17)	Increase (n = 9)	*P* Value
Mechanical ventilation, h	158.3 [71–335.4]	125.1 [67.3–313.5]	233.4 [82.6–358.7]	149.5 [68.2–317.7]	0.506
Ventilator-free days (28 days)	9 [0–24]	10 [0–25]	0 [0–22]	0 [0–20]	0.482
Tracheostomy, n (%)	9 (12)	7 (14)	2 (11)	0 (0)	0.483
Reconnected to mechanical ventilation < 7 days after extubation, n (%)	12 (16)	8 (16)	2 (11)	2 (22)	0.748
Length of ICU stay, days	9 [5–21]	8 [5–22]	10 [7–19]	7 [4–23]	0.676
Length of hospital stay, days	21 [9–36]	22 [8.8–36]	15 [11–36]	26 [5–35]	0.975
ICU mortality, n (%)	31 (40)	18 (36)	9 (50)	4 (44)	0.562
Hospital mortality, n (%)	34 (44)	19 (38)	10 (56)	5 (56)	0.334

Data are expressed as median [interquartile range] or n (%).
ICU, intensive care unit.

system compliance, intrinsic PEEP, or low inspiratory muscle capacity as is common in ICU patients.[1]

Reproducibility of Expiratory Muscle Ultrasound and the Effect of Lung Volume

In the current study, we demonstrate that ultrasound of the expiratory muscles is feasible and highly reproducible in ICU patients. The probe position on the skin was marked to reduce variability in repeated measurements originating from muscle anatomy.[17] The repeatability coefficient for thickness measurements of the expiratory muscles ranged from 1.0 to 1.6 mm, which is higher than the range of 0.2 to 0.4 mm reported for the diaphragm.[17] The origin of this difference is unknown but might be related to the echogenic properties of the muscles (as probes with identical properties were used). Because the expiratory muscles are much thicker than the diaphragm (13.2 ± 3.9 mm *versus* 2.4 ± 0.8 mm), the ratio of measurement variance to the thickness of the muscles is equal to 10% in both muscle groups.

Increasing lung volume (passive or active) will result in caudal movement of the diaphragm, which in turn is expected to stretch the abdominal wall muscles. Indeed, the thickness of the expiratory muscles significantly decreased with tidal inspiration. The magnitude of this difference (± 3% with tidal volume of ± 480 ml) was, however, much smaller than the difference that was used to categorize patients (15%). It is therefore unlikely that the differences in expiratory muscle thickness observed in the cohort study are explained by differences in lung volume. An earlier study demonstrated that in healthy subjects, breathing from functional residual capacity to residual volume significantly increased expiratory muscle thickness.[32] However, as subjects performed an expiratory maneuver, the increase in thickness in that study was at least partly explained by active muscle contraction.

Loss of Muscle Thickness

This study used ultrasound to assess whether thickness changes of the expiratory muscles would occur in patients during the first week of mechanical ventilation. The development of expiratory muscle atrophy in critically ill patients has been observed in rectus abdominis muscle biopsies.[11] *In vitro* contractility of the rectus abdominis muscle was reduced in septic mechanically ventilated patients.[33] Studies using muscle biopsies are important to identify biochemical pathways involved in the development of atrophy but cannot be used to study time-dependent changes in muscle thickness. Ultrasound provides a more comprehensive overview of the changes in the different muscle groups of the respiratory pump, and it is a feasible technique to study the time-dependent changes in thickness. Of our patients, 22% developed atrophy of the expiratory muscles. These patients could not be identified by specific clinical or physiologic characteristics at baseline. Previous studies have demonstrated that the presence of expiratory muscle weakness in critically ill patients is associated with adverse clinical outcome. Reduced thickness of the expiratory muscles may impair strength and as such negatively affect airway clearance, resulting in atelectasis and pneumonia, especially after extubation.[22,23,25] In our sensitivity analyses, we found that patients who developed expiratory muscle atrophy had significantly longer hospital length of stay, but this finding should be interpreted with caution because this study was not designed to investigate the functional implications of expiratory muscle atrophy. This hypothesis remains to be investigated in a larger sample size, and the data from this study are useful for sample size calculation.

Increased Muscle Thickness

Previous studies reported increased diaphragm thickness in a subgroup of ventilated critically ill patients.[15] In this study, we found that thickness of the expiratory muscles increased in 12% of patients, but interestingly, this increase in thickness was mainly driven by increased thickness of the interparietal fasciae between the three muscle layers. This is a novel and unexpected finding. The muscular

fascia is a highly organized connective tissue containing different types of cells (*e.g.*, fibroblasts, myofibroblasts) and extracellular matrix molecules (*e.g.*, ground substance and collagen fibers).[34] This fascia plays a crucial role in transmission, distribution, and absorption of muscle force.[34,35] Both mechanical unloading and loading affect connective tissue collagen synthesis and degradation, thus leading to fascia tissue remodeling.[34,36] This activity-driven adaptation may play a role in regulation of muscle mass and strength.[37,38] However, excessive or repetitive loading to fascial tissue initiates persistent inflammation, inducing macrophages and cytotoxic levels of cytokines, ultimately resulting in tissue damage.[39] Cytokines, such as interleukins, tumor necrosis factor α, and transforming growth factor β, are fibrogenic cytokines facilitating fibrosis (*e.g.*, fibroblast proliferation and collagen matrix deposition).[34,40,41] Morphological characteristics and function of fascia tissue in respiratory muscles of critically ill patients have not been studied. Future biopsy studies should further elucidate the role of the fasciae in regulating muscle mass and function in ventilated patients and evaluate the clinical implications of increased respiratory muscle fascia thickness.

Association between Expiratory Muscles Atrophy and Diaphragm Atrophy

Another important finding of the current study is that we found no significant association between thickness changes in the expiratory muscles and the diaphragm. The fact that the diaphragm and the expiratory muscles respond differently to mechanical ventilation and critical illness is remarkable, because both muscles act in the same metabolic environment (*e.g.*, level of systemic inflammation, reactive oxygen/nitrogen species, drug exposure, arterial oxygen tension, pH). However, these findings are consistent with earlier studies from our group and others demonstrating that critical illness does not equally affect the different muscles of the respiratory pump.[10,12] For instance, muscle biopsy studies in ICU patients demonstrated different severities of atrophy between the diaphragm and noninspiratory muscles (mostly rectus abdominis muscle).[12]

Strengths and Limitations

The strengths of the current study include the relatively large sample size, simultaneous analysis of different muscles of the respiratory pump, and separate analysis of the muscular fasciae. Several limitations should be acknowledged. First, this is a single-center study conducted in severely ill ventilated patients (hospital mortality ± 44%). Whether similar results are obtained in other patient categories remains to be investigated. Nevertheless, this study was performed in a large academic ICU admitting a heterogeneous group of ICU patients. Second, we could not evaluate the effects of mechanical ventilation *per se* on the expiratory muscles but merely the combined effects of critical illness and mechanical ventilation. To precisely investigate the contribution of mechanical ventilation, a nonventilated control group with similar severity of disease should be included. This was not feasible, and therefore the exact contribution of mechanical ventilation should be interpreted with caution. Another limitation of the study is early extubation in many patients, because only one third of the patients remained ventilated by the end of the first week. This might lead to biased observations, because the least-ill and most-ill patients tend to drop out earlier. This form of bias is inherent to cohorts of ICU patients, but it does mean that our results should be interpreted with caution. Further studies are required to confirm the pattern of thickness changes of the expiratory muscles that we have observed and to assess the functional implications.

Conclusions

The current study demonstrates that ultrasound is a highly reliable tool to assess expiratory muscle thickness in mechanically ventilated critically ill patients. Tidal volume has a significant although small effect on expiratory muscle thickness. Atrophy develops in 22% of the patients and is attributable to loss of muscle tissue; increased expiratory muscle thickness develops in 12% of the patients and is attributed to increased thickness of the interparietal fasciae. Changes in thickness of the expiratory muscles are not associated with changes in diaphragm muscle thickness, indicating that different muscles of the respiratory pump may respond differently to critical illness and mechanical ventilation.

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Competing Interests

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