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Ultrasonographic Assessment of the Diaphragm in Chronic Obstructive Pulmonary Disease Patients: Relationships with Pulmonary Function and the Influence of Body Composition – A Pilot Study

Andrea Smargiassi Riccardo Inchingolo Linda Tagliaboschi
Alessandro Di Marco Berardino Salvatore Valente Giuseppe Maria Corbo

Pulmonary Medicine Department, Università Cattolica del Sacro Cuore, Rome, Italy

For editorial comment see p. 355

Key Words
Chronic obstructive pulmonary disease · Diaphragm · Chest ultrasound

Abstract
Background: Skeletal muscle weakness with loss of fat-free mass (FFM) is one of the main systemic effects of chronic obstructive pulmonary disease (COPD). The diaphragm is also involved, leading to disadvantageous conditions and poor contractile capacities. ***Objectives:*** We measured the thickness of the diaphragm (TD) by ultrasonography to evaluate the relationships between echographic measurements, parameters of respiratory function and body composition data. ***Methods:*** Thirty-two patients (23 males) underwent (1) pulmonary function tests, (2) echographic assessment of TD in the zone of apposition at various lung volumes, i.e. TD at residual volume (TDRV), TD at functional residual capacity (TDFRC) and TD at total lung capacity (TDTLC), and (3) bioelectrical body impedance analysis. The BMI and the BODE (BMI-Obstruction-Dyspnea-Exercise) index values were reported. ***Results:*** TDRV, TDFRC and TDTLC measured 3.3, 3.6 and 6 mm, respectively, with good intraobserver reproducibility (0.97, 0.97 and 0.96, respectively). All the TDs were found to be related to FFM, with the relationship being greater for TDFRC ($r^2 = 0.39$ and $p = 0.0002$). With regard to lung volumes, inspiratory capacity (IC) was found to be closely related to TDTLC ($r^2 = 0.42$ and $p = 0.0001$). The difference between TDTLC and TDRV, as a thickening value (TDTLCRV), was closely related to FVC ($r^2 = 0.34$ and $p = 0.0004$) and to air-trapping indices (RV/TLC, FRC/TLC and IC/TLC): the degree of lung hyperinflation was greater and the TDTLCRV was less. Finally, we found a progressive reduction of both thicknesses and thickenings as the severity of IC/TLC increased, with a significant p value for the trend in both analyses ($p = 0.02$). ***Conclusions:*** Ultrasonographic assessment of the diaphragm could be a useful tool for studying disease progression in COPD patients, in terms of lung hyperinflation and the loss of FFM.

Introduction

Chronic obstructive pulmonary disease (COPD) is a progressive disease characterized by an incomplete reversible airflow limitation.

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Riccardo Inchingolo, MD
Pulmonary Medicine Department
Università Cattolica del Sacro Cuore, Largo Gemelli 8
IT–00168 Roma (Italy)
E-Mail r_inchingolo@virgilio.it

The disease is determined by the manifestation of chronic inflammation with acute exacerbations, alterations in repair mechanisms, remodeling, oxidative stress imbalance and 'spill-over' of inflammatory mediators into the circulation, resulting in important systemic manifestations [1].

Systemic inflammation can initiate or worsen comorbid diseases such as ischemic heart disease, heart failure, osteoporosis, normocytic anemia, lung cancer, depression and diabetes. These diseases potentiate the morbidity of COPD, leading to increased hospitalization, mortality and healthcare costs, and complicating the management of COPD patients [1].

Systemic inflammation, deconditioning, oxidative stress, nutritional imbalance, cardiac failure, hypoxemia and hypercapnia can also lead to a loss of fat-free mass (FFM) [2] and to weakening of the skeletal muscles, considered to be the main systemic effects influencing the progression of the disease [3, 4]. Long-term administration of steroids (even low doses), used widely in the treatment of COPD, can also contribute to respiratory muscle weakness with wasting of the diaphragm muscle [5, 6].

Several studies have shown that the function and structure of skeletal muscle are altered in COPD patients and these abnormalities are related to respiratory function, exercise intolerance, health status, mortality and the utilization of healthcare resources [7–9]. Muscle wasting is associated with a loss of muscle strength and has therefore been identified as a significant determinant of mortality in COPD, independent of lung function, smoking and BMI [7–9].

It has been demonstrated that FFM is more accurate than BMI for predicting COPD severity, as it is related not only to the distance walked in the 6-minute walk test (6MWT) but also to the FEV_1/FVC ratio, the Modified Medical Research Council (MMRC) dyspnea scale and the percentage of predicted FEV_1 [10].

The diaphragm is the main respiratory muscle. Patients affected by emphysematous COPD with a loss of FFM and muscle wasting show profound alterations regarding the mass, thickness and area of the diaphragm [11–13].

The study of the diaphragm is thus considered a key point in the evaluation of patients with severe COPD and several methods are employed such as magnetic resonance scans [14], phrenic-nerve conduction study [15] and invasive assessment of transdiaphragmatic pressure (the difference between the intrathoracic and intraabdominal pressure during the respiratory cycle) [16].

Recently, the use of ultrasonographic (US) techniques for the assessment of both diaphragmatic excursions [17, 18] and thickness of the diaphragm (TD) at different lung volumes [19] has been proposed. The reproducibility of measurements and the relationships between diaphragm kinetics and respiratory functional parameters have been investigated in normal subjects.

TD is also related to muscle mass. Recently, in patients with cystic fibrosis, TD has been found to be related to both lung volume and FFM [20].

In COPD patients, obtaining a noninvasive evaluation of the diaphragmatic function from US measurements of thickness, thickening and excursions could be a useful way to estimate the severity of the disease.

The aim of our study was to investigate, in a sample of COPD patients, the US features of the diaphragm (i.e. thickness, thickening and excursions) in relation to both body composition (in terms of FFM) and pulmonary function.

Materials and Methods

This protocol received the approval of the Ethical Committee of the Catholic University of the Sacred Heart, Rome (No. P/498/CE/2011), and every patient gave her/his written informed consent.

Study Design

The study included COPD patients from April 2011.

The diagnosis of COPD was based on an investigation of their medical history, a clinical examination and respiratory function tests [21].

Patients were included who had an FEV_1/FVC ratio of <70%, irreversible after the administration of beta2 agonists (reversibility test). Patients affected by neurological disease, lung cancer, pleural effusion, chemical pleurodesis, interstitial lung disease or with a pacemaker device or a medical history of major surgical chest/abdominal intervention were excluded.

The patients underwent respiratory function tests, echographic measurements and bioelectrical impedance analysis.

Methods

Pulmonary Function Tests

Each patient underwent pulmonary function tests using the technique of body plethysmography (Platinum Elite Series™, Medical Graphics Corp., Milan, Italy).

These tests included simple spirometry with the measurement of pulmonary slow and forced flows, lung volumes and airways resistance, reversibility with beta2 agonists and lung diffusion of carbon monoxide (DLCO). Respiratory function tests were performed according to other studies [22–24].

Three reproducible maneuvers were obtained for each test and the best one was reported.

Finally, each patient underwent a 6MWT [25]. The distance walked, the arterial oxygen saturation of hemoglobin (measured by pulse oximeter at the beginning and end of the exercise) and the

Table 1. Descriptive characteristics of the study population

	Mean	Standard deviation	Median
Age, years	71.8	8.32	72
FEV_1, liters	1.25	0.43	1.23
FEV_1, %	50.6	16.3	47
FVC, liters	2.6	0.81	2.48
FVC, %	81.1	20.9	78.5
VC, liters	3.02	0.85	3.02
VC, %	89.7	19.8	88
FEV_1/FVC	0.48	0.1	0.48
DLCO/VA	2.92	0.97	2.86
DLCO/VA, %	66.5	22.5	65
TLC, liters	6.68	1.37	6.59
TLC, %	112	21	109
FRC, liters	4.3	1.15	4.05
IC, liters	2.37	0.74	2.42
RV, liters	3.69	1.17	3.41
RV, %	152	48.5	149
FRC/TLC	0.64	0.08	0.62
IC/TLC	0.36	0.08	0.38
RV/TLC	0.54	0.1	0.57
Distance walked, m	387.76	125.9	400
BODE index value	2.9	1.95	3
BMI	27.6	5.2	27.5
Height, m	1.67	0.01	1.68
FFM, kg	53.3	9.2	55.5

VA = Alveolar volume.

degree of dyspnea estimated according to the MMRC scale [26] were reported. The collection of these parameters allowed calculation of the BODE (BMI-Obstruction-Dyspnea-Exercise) index score [27] for each patient.

Echographic Measurements

Echographic measurements were performed using a MyLab™ 50 Gold Cardiovascular (Esaote Spa, Rome, Italy) ultrasound machine with probes of 3.5–5 and 7.5–12 MHz.

Each patient was asked to lie in a semirecumbent position (i.e. a bed slope of 45°).

The excursion of the right hemidiaphragm during maximal profound inspiration starting from normal end-expiratory volume (functional residual capacity, FRC) was obtained by means of a US technique reported by Testa et al. [18].

We performed an examination using a convex probe of 4 MHz.

Measurements of TD and diaphragm kinetics at the zone of apposition were performed using a technique similar to that presented by Ueki et al. [19]. TD was measured at the closest point to the 'curtain sign' in which the two hyperecogenic parallel layers of the diaphragm were clearly identified in the right intercostal position.

We performed the examination by means of intercostal scanning, using a linear probe of 10 MHz, with the patient lying in a semirecumbent position. The TD measurements were made at the end of a normal expiration (corresponding to FRC), during a breath-holding maneuver after maximal inspiration (corresponding to total lung capacity, TLC) and at the end of maximal expiration (corresponding to residual volume, RV), and they were reported as TDFRC, TDTLC and TDRV, respectively. Two measurements were performed by the same operator (A.S.) blinded to the result, and the mean value was then reported.

The differences between TDTLC and TDFRC (TDTLCFRC) and TDTLC and TDRV (TDTLCRV) were reported as diaphragm thickenings.

Bioelectrical Impedance Analyses

Bioelectrical body impedance assessment was performed by means of a Tanita SC-240 MA impedentiometer (Biologica Tecnologia Médica SL, Barcelona, Spain). Bioelectrical impedance analysis is a method for estimating body composition by introducing the weight, height and waist circumference of each subject from the determined impedance a number of parameters [28–30].

The FFM for each patient was also reported.

Analysis

Functional parameters, body composition data and echographic parameters were reported as means and standard deviations.

The reproducibility of the echographic measurements was calculated by the reproducibility coefficient of Bland and Altman [31]. The intraclass analysis of correlation, reporting the intraclass R factor, was performed to define the significance. ANOVA was used for comparison analysis.

The relationships between functional, echographic and body composition variables were analyzed by means of linear and multiple regression models. Firstly, we analyzed the relation between TD and static lung volume (TLC, FRC, IC and RV), their ratios (RV/TLC, IC/TLC and FRC/TLC) and FFM separately. To define the determinant factors of TDTLC, a multiple regression was performed including TDTLC as an independent parameter and lung volume and FFM as dependent variables. Secondly, the relation between diaphragm thickening with dynamic volumes, such as FVC and FEV_1, static volumes, ratios and FFM was evaluated. The same analysis was performed as that for diaphragmatic excursion. Lastly, to define the determinant factors of TDTLCRV, a multiple regression was performed.

In order to better evaluate the effects of air trapping on TD and diaphragm thickening, we divided COPD patients into 3 groups, based on the frequency distribution of the variable IC/TLC, each group corresponding to a tertile of the distribution: score 0 = IC/TLC >0.40 (group 0), score 1 = IC/TLC between 0.40 and 0.31 (group 1) and score 2 = IC/TLC <0.31 (group 2). The mean values of thicknesses and thickenings were calculated for each group; they were adjusted for FFM and a test for trend was applied.

Statistical significance was considered for $p < 0.05$.

Results

Descriptive Statistics

Thirty-two COPD patients (23 males) were enrolled in this study. The mean BMI was 27.6 ± 5.2 with a minimum value of 17.2 and a maximum value of 38.8, ranging from underweight to subjects affected by 2nd-degree obesity.

Table 1 shows the descriptive characteristics of the study population.

The respiratory function tests showed patients belonging to every COPD severity grade [32] with high levels of air trapping: mild (n = 3), moderate (n = 11), severe (n = 14) and very severe (n = 4). The median BODE index value was 3 with a maximum value of 7 and a minimum value of 0.

We reported high intraoperator reproducibility for the echographic measurements (table 2).

The measurements were performed twice by the same operator and the mean value was reported for subsequent statistical analysis.

The diaphragmatic excursion and TD are shown in table 3.

TD changed according to lung volumes increasing slightly, passing from RV to FRC and sharply from FRC to TLC (fig. 1). TD was significantly greater in males than in females ($p < 0.001$).

TD, FFM and Lung Volume

TD at different lung volumes was closely related to IC, vital capacity (VC) and TLC, showing TDTLC to have the closest relationships with IC and VC ($r^2 = 0.42$ and $p = 0.0001$ in both cases). Moreover, TDTLC was related to all indices of air trapping: directly to IC/TLC ($p = 0.01$), inversely to FRC/TLC and to RV/TLC ($p = 0.01$ and $p = 0.02$, respectively; table 4).

FFM showed significant direct correlations with all TD measurements (i.e. TDFRC, TDTLC and TDRV), with closer relationships with TDFRC and TDTLC ($r^2 = 0.39$ and $p = 0.0002$ and $r^2 = 0.32$ and $p = 0.0008$, respectively). Lastly, all thicknesses were found to be related to physical characteristics like height and BMI (table 4). The multiple regression analysis demonstrated that the best model ($r^2 = 0.58$) investigating TDTLC included IC ($p = 0.0003$) and FFM ($p = 0.011$) as determinant factors.

Thickenings and Excursion

Both TDTLCFRC and TDTLCRV were closely related to VC, FVC and IC. TDTLCRV and TDTLCFRC were related to air-trapping indices (RV/TLC, FRC/TLC and IC/TLC): the degree of lung hyperinflation was greater and that of TDTLCRV and TDTLCFRC was less. Moreover, a close relationship was found between diaphragm thickenings and FEV_1 (table 5).

Diaphragm excursion did not show any relationship with the parameters of respiratory function but was found to be closely related to BMI ($r^2 = 0.29$ and $p = 0.001$) (table 5).

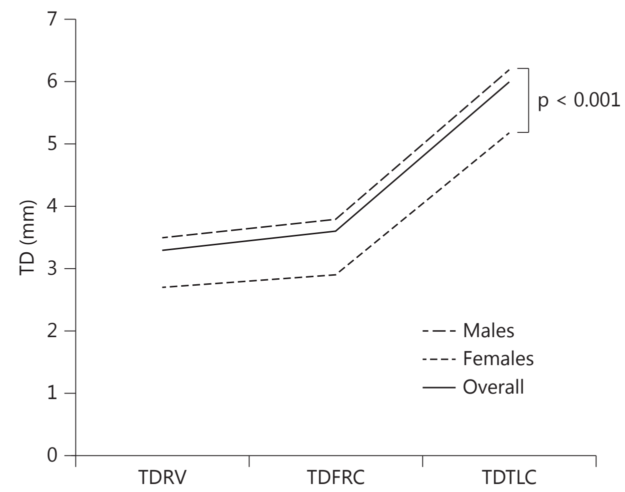

Fig. 1. TD changes according to lung volumes. TDs are significantly greater in males than in females.

Table 2. Intraoperator reproducibility of diaphragm measurements

	Mean 1	Mean 2	Mean	Coefficient of reproducibility	Ri
Excursion	6.0	6.2	6.1	1.8	0.90
TDRV	3.3	3.3	3.3	0.3	0.97
TDFRC	3.6	3.6	3.6	0.4	0.97
TDTLC	5.9	6.1	6.0	0.7	0.96

Table 3. Diaphragm echographic measurements

	Mean	Standard deviation	Median
Excursion, cm	6.1	1.98	6.2
TDRV, mm	3.3	0.66	3.3
TDFRC, mm	3.6	0.71	3.7
TDTLC, mm	6.0	0.96	6.1

TDTLCRV and TDTLCFRC did not show any relationship with body composition data.

The multiple regression analysis demonstrated that the best model ($r^2 = 0.58$) investigating TDTLCRV included IC/TLC ($p = 0.0001$) and FEV_1/FVC ($p = 0.008$) as determinant factors.

Table 4. Correlations between TDs at different lung volumes and lung volumes, ratios, FFM and BMI

	TDFRC			TDTLC			TDRV		
	beta	r^2	p	beta	r^2	p	beta	r^2	p
VC, liters	0.39	0.21	0.008	0.74	0.42	0.0001	0.28	0.13	0.04
IC, liters	0.41	0.18	0.01	0.84	0.42	0.0001	0.39	0.19	0.01
RV, liters	0.15	0.06	NS	0.006	0.0001	NS	0.16	0.08	NS
FRC, liters	0.20	0.10	0.06	0.06	0.005	NS	0.17	0.09	0.09
TLC, liters	0.26	0.24	0.004	0.28	0.16	0.02	0.23	0.22	0.005
RV/TLC	–0.90	0.01	NS	–3.55	0.15	0.02	–0.23	0.001	NS
FRC/TLC	–0.98	0.01	NS	–4.72	0.18	0.01	–0.99	0.01	NS
IC/TLC	0.98	0.01	NS	4.72	0.18	0.01	0.99	0.01	NS
FFM, kg	0.05	0.39	0.0002	0.06	0.32	0.0008	0.04	0.30	0.0012
BODE index value	–0.03	0.01	NS	–0.07	0.02	NS	–0.03	0.008	NS
BMI	0.06	0.21	0.0078	0.07	0.16	0.021	0.06	0.24	0.0039
Height, m	5.31	0.36	0.0003	5.81	0.24	0.005	3.76	0.21	0.08

Table 5. Relationships between diaphragm thickenings and excursion with lung volumes, ratios, FFM and BMI

	TDTLCRV			TDTLCFRC			Excursion		
	beta	r^2	p	beta	r^2	p	beta	r^2	p
VC, liters	0.46	0.34	0.0004	0.35	0.23	0.005	0.33	0.02	NS
FVC, liters	0.48	0.34	0.0004	0.37	0.24	0.004	0.12	0.003	NS
IC, liters	0.44	0.26	0.002	0.42	0.26	0.002	0.85	0.1	NS
FEV_1	0.70	0.21	0.007	0.60	0.18	0.01	0.74	0.02	NS
FEV_1/FVC	–0.63	0.01	NS	–0.31	0.003	NS	2.95	0.02	NS
RV, liters	–0.16	0.08	NS	–0.15	0.08	NS	0.32	0.03	NS
FRC, liters	–0.11	0.04	NS	–0.14	0.07	NS	0.18	0.01	NS
TLC, liters	0.05	0.01	NS	0.02	0.003	NS	0.37	0.06	NS
RV/TLC	–3.32	0.29	0.001	–2.65	0.21	0.007	0.27	0.0002	NS
IC/TLC	3.73	0.24	0.003	3.74	0.28	0.001	4.3	0.03	NS
FRC/TLC	–3.73	0.24	0.003	–3.74	0.28	0.001	–4.3	0.03	NS
FFM, kg	0.02	0.076	NS	0.01	0.03	NS	0.07	0.11	0.06
BODE index value	–0.04	0.02	NS	–0.04	0.01	NS	–0.03	0.001	NS
BMI	0.01	0.09	NS	0.01	0.09	NS	0.2	0.29	0.001
Height, m	2.06	0.06	NS	0.50	0.004	NS	3.1	0.016	NS

Role of IC/TLC Values on Thickness and Thickening

Figure 2 shows the TDTLC and TDTLCRV mean values adjusted for FFM for each group. There was a progressive reduction in both thickness and thickening, increasing the severity of air trapping with a significant p for the trend in both analyses (p = 0.02).

Finally, in our sample, IC/TLC was found to be strongly related to the BODE index value (r^2 = 0.44 and p = 0.0003) and the distance walked in the 6MWT (r^2 = 0.30 and p = 0.002).

Discussion

Our results demonstrate that TD measured with the US technique in COPD patients was related to lung volume. Moreover, both FFM and BMI were found to have a close association. Indices of lung hyperinflation were found to be related only with regard to the measurement of TDTLC. Diaphragm thickening was related to the indices of lung hyperinflation and dynamic volumes such as VC, FVC and FEV_1.

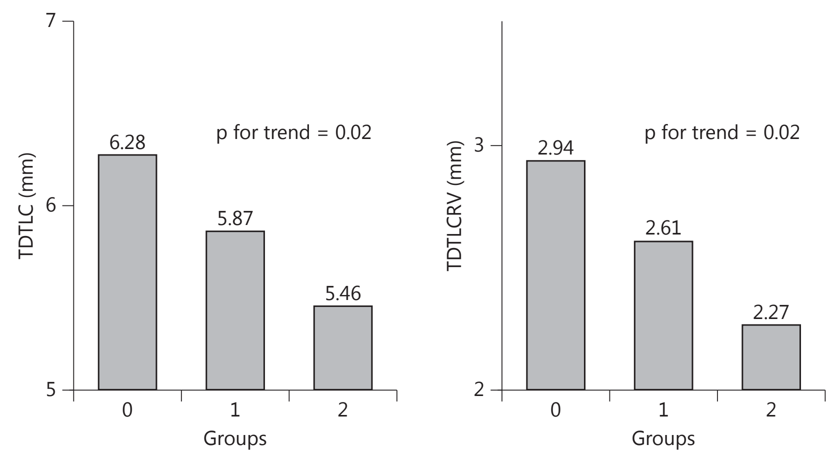

Fig. 2. TDTLC and TDTLCRV mean values adjusted for FFM for each group. There is a progressive reduction of both thicknesses and thickenings, increasing the severity of air trapping.

In recent years, the use of ultrasound has gained wide acceptance, also in pneumological settings. However, the US characteristics of respiratory muscles in COPD patients, particularly the diaphragm, have not been fully studied.

Using standard techniques [18, 19], the measurement is feasible (perhaps with the exception of severe obesity due to the high acoustic impedance and the thickness of subcutaneous tissues) and reproducible. In our study, we calculated the intraoperator reproducibility and reported a very high level of agreement of both TDs and diaphragm kinetics (table 2), which confirmed previous data on healthy subjects [17, 18, 33].

According to Cohn et al. [33], we showed that the diaphragm thickened as the lung volume increased, from RV to TLC, with a more rapid rate of thickening at the highest lung volume and a significantly greater thickening in males than in females.

Diaphragm contraction is maximal at full inspiration, so the TDTLC could be considered as an indicator of the contractile capacities of this muscle. These contractile capacities could be limited by both lung hyperinflation and muscle wasting under both these conditions in COPD patients. We found that TDTLC was related to the indices of lung hyperinflation, FFM and BMI. In the multiple regression analysis, the IC and FFM were found to be the main determinants of TDTLC. On the other hand, when the diaphragm relaxed (at FRC), FFM was found to be the main determinant of the TD and there was no association with the indices of lung hyperinflation.

With regard to diaphragmatic thickenings, both TDTLCRV and TDTLCFRC were found to be related to air trapping, hyperinflation indices and dynamic pulmonary volumes (VC, FVC and FEV_1). No correlation with FFM and BMI was found. Thus, in the dynamic process of thickening, the main role seemed to be played by airflow limitation resulting in air trapping and lung hyperinflation: FEV_1/FVC and IC/TLC were found to be determinants of TDTLCRV.

Previous papers have stated that IC/TLC is a crucial functional parameter that is related to the BODE index value and the distance walked in the 6MWT as well as being a predictor of mortality [34, 35]. As expected, in our sample, IC/TLC was found to be strongly related to the BODE index value and the distance walked in the 6MWT.

The important role of IC/TLC on TD and thickening at TLC (TDTLC and TDTLCRV) can be demonstrated by considering the 3 groups previously described. When TDTLC and TDTLCRV mean values for each group were adjusted for FFM, a significant trend was found: patients with a higher degree of air trapping had a lower TD and thickening at TLC volume (TDTLC and TDTLCRV).

Diaphragmatic excursion must be considered separately. According to some of the authors [18, 36] of studies on healthy subjects, no significant relationships were found between the right hemidiaphragmatic excursion and the respiratory function parameters. Others found significant correlations with the degree of air trapping, TLC and FEV_1/FVC ratio [37], although a different technique was used to establish the excursion. We found that diaphragmatic excursion showed direct significant correlations only with BMI.

Some limitations of the study should be mentioned. Lung flow and volume were measured with the patients

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in an orthostatic position and the echographic measurements were performed holding the patients in a semirecumbent position, according to a technique described by Testa et al. [18]. In this setting, the role played by the abdomen and the abdominal muscles can differ, leading to a reduction of the total inspired volume due to increased air trapping and to a change in the end expiratory volume. Correlations of diaphragm measurements and lung volumes could be affected by this bias. However, we decided to use this technique because we wanted to test the relationships of measurements performed under different conditions, i.e. even in patients who cannot maintain the orthostatic position. The echographic technique of acquisition of TD was therefore quite different from the technique already described by Ueki et al. [19]. We first measured the TD in COPD patients at the closest point to the 'curtain sign' both at FRC and at TLC. Moreover, the patients in our sample were not seated but were in a semirecumbent position. These conditions may modify the echographic characteristics of the diaphragm in the zone of apposition and the behavior of the diaphragm during contraction, especially in hyperinflated COPD subjects.

Another limitation is the mean low-severity score (BODE index) of our sample, with a mortality rate that was significantly greater for a BODE index value of >6. Therefore, at the present time, we cannot consider TDs and diaphragm thickenings as prognostic parameters of mortality.

Finally, in our study, only the intraobserver reproducibility was studied, with a high level of agreement. However, in ultrasonography, interobserver agreement is an important issue and this pilot study had the intention of stimulating further studies in order to assess the interobserver reproducibility of TDs in COPD patients.

In conclusion, we suggest that echographic measurements of TDTLC and thickening at TLC might be a useful tool to estimate lung hyperinflation, especially when adjusted for FFM. Moreover, this technique could be feasible in patients in critical clinical settings or those on mechanical ventilation, and facilitate the acquisition of data on diaphragm performance.

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