



ERS statement on respiratory muscle testing at rest and during exercise

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Diverse methods are available for assessment of the respiratory muscles; the technique used should be tailored to the question posed <http://ow.ly/poE830o8y58>

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ABSTRACT Assessing respiratory mechanics and muscle function is critical for both clinical practice and research purposes. Several methodological developments over the past two decades have enhanced our understanding of respiratory muscle function and responses to interventions across the spectrum of health and disease. They are especially useful in diagnosing, phenotyping and assessing treatment efficacy in patients with respiratory symptoms and neuromuscular diseases. Considerable research has been undertaken over the past 17 years, since the publication of the previous American Thoracic Society (ATS)/European Respiratory Society (ERS) statement on respiratory muscle testing in 2002. Key advances have been made in the field of mechanics of breathing, respiratory muscle neurophysiology (electromyography, electroencephalography and transcranial magnetic stimulation) and on respiratory muscle imaging (ultrasound, optoelectronic plethysmography and structured light plethysmography). Accordingly, this ERS task force reviewed the field of respiratory muscle testing in health and disease, with particular reference to data obtained since the previous ATS/ERS statement. It summarises the most recent scientific and methodological developments regarding respiratory mechanics and respiratory muscle assessment by addressing the validity, precision, reproducibility, prognostic value and responsiveness to interventions of various methods. A particular emphasis is placed on assessment during exercise, which is a useful condition to stress the respiratory system.

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Introduction	3
Methods	3
Section 1. Respiratory muscle function	4
<i>Andrea Albuquerque, Tony Babb, Martin Dres, Brigitte Fauroux, Jordan A. Guenette, Hans Joachim Kabitz, Franco Laghi, Daniel Langer, Pierantonio Laveneziana, J. Alberto Neder, Denis O'Donnell, Mickael I. Polkey, Andrea Rossi, Christina M. Spengler, Samuel Verges</i>	
1.1. Airway opening, oesophageal and gastric pressures: technical considerations	4
1.1.1. Pressure measurement	4
1.1.2. Pressure assessment devices	4
1.1.2.1. Pressure transducers	4
1.1.2.2. Probes for invasive pressure assessment	5
1.1.2.3. Devices for measurement of airway opening pressure	5
1.2. Voluntary tests of respiratory muscle strength	5
1.2.1. Maximal static inspiratory and expiratory mouth pressure	5
1.2.2. Maximal sniff nasal inspiratory pressure	6
1.2.3. Peak cough flow	6
1.3. Voluntary manoeuvres with oesophageal and gastric pressures	7
1.4. Respiratory muscle-related mechanics of breathing	7
1.4.1. Lung function testing	7
1.4.2. Indices of respiratory muscle effort	7
1.5. Evoked manoeuvres	10
1.6. Respiratory muscle endurance testing	11
1.6.1. Maximal incremental load testing	12
1.6.2. Constant load testing	12
1.6.3. Time trial	12
Section 2. Respiratory muscle neurophysiology	12
<i>Martin Dres, Anna L. Hudson, Franco Laghi, Yuan-Ming Luo, Andrea Rossi, Frédéric Series, Thomas Similowski</i>	
2.1. Electromyography	12
2.2. Electroencephalography	13
2.3. Transcranial magnetic stimulation	16
Section 3. Respiratory muscle imaging	19
<i>Andrea Aliverti, Martin Dres, Bruno-Pierre Dubé, Brigitte Fauroux, Franco Laghi, Andrea Rossi</i>	
3.1. Ultrasound	19
3.1.1. Diaphragm thickness	19
3.1.2. Diaphragm thickening fraction and ratio	19
3.1.3. Diaphragm excursion	20
3.2. Optoelectronic plethysmography	20
3.3. Other investigations	20
Section 4. Respiratory muscle structure, perfusion and metabolism	21
<i>Esther Barreiro, Joachim Gea, J. Alberto Neder, Roberto A. Rabinovich, Ioannis Vogiatzis</i>	
4.1. Near-infrared spectroscopy	21
4.2. Oxygen cost of breathing	22
4.3. Biopsy (specificities for respiratory muscles)	22
4.4. Typology	23
4.5. Mitochondrial function	23
4.6. Oxidative stress	24
4.7. Inflammation	24
Conclusion	24
References	25

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Introduction

Assessing respiratory mechanics and respiratory muscle structure and function is an essential component of both clinical practice and research. It is especially useful in patients with respiratory symptoms and neuromuscular diseases (NMDs), contributing to diagnosis, patient phenotyping, assessment of treatment efficiency and patient follow-up. The American Thoracic Society (ATS) and the European Respiratory Society (ERS) published a statement on respiratory muscle testing in 2002, reviewing the rationale and technical characteristics of the main methods available [1]. Nearly two decades later, given the large amount of novel research in the field, the chairs of the present task force felt a need to summarise the latest knowledge on respiratory mechanics and muscle assessment both for clinicians and researchers. Since 2002, key advances have been made in the field of mechanics of breathing, respiratory muscle neurophysiology, and respiratory muscle imaging in health and disease, including in paediatrics and critically ill patients in the intensive care unit (ICU). A specific focus of the task force has been the assessment of respirator muscles and mechanics during exercise, a situation stressing the respiratory system and thus allowing the evaluation of respiratory muscle response to increased ventilatory demand.

Methods

The task force was formed in June 2016, composed of experts from the ERS Clinical Respiratory Physiology, Exercise and Functional Imaging Group (04.01), the ERS Rehabilitation and Chronic Care Group (01.02), the Physiotherapists Group (09.02), and representatives from the European Lung Foundation and the ERS Science Council. The task force received support from ERS methodologists throughout the project. Three meetings of the task force were held; two during the annual congress of the ERS (September 2016 and 2017) and one in Lausanne in March 2017. All task force members signed conflict of interest disclosures at the beginning of the project and updated them at project finalisation or when any new relevant conflict of interest appeared. Conflicts of interest were managed according to ERS rules.

Studies that reported the evaluation of respiratory muscles (inspiratory and expiratory) and upper airway muscles at rest or during exercise in adults and children with cardiorespiratory diseases were reviewed, without restrictions on study design. MEDLINE and Cochrane Library records from 1970 to 2017 were searched. Selected references considered to be of particular relevance were included up to June 2018. Reference lists of all primary studies and review articles were examined for additional citations. Only studies written in English, or for which an English translation was available, were consulted. Studies were included that refer (singly or in combination) to reported validity (*i.e.* the extent to which a test or variable is related to the function of a physiological system or to patient-meaningful variables, such as symptoms or exercise), precision or reproducibility, prognostic information (*i.e.* relationship with the natural history of the disease), discrimination (*i.e.* whether a variable can differentiate the severity of the disease as conventionally measured), clinical meaningful difference (*i.e.* the minimal difference in a tested variable that is considered to be functionally worthwhile or clinically important), or test response to interventions. Studies that did not meet the inclusion criteria based on title or abstract were excluded.

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Studies that met the inclusion criteria were retrieved in full text to determine whether they were suitable for inclusion. For each section, the articles selected by the primary task force author had to be approved by a second author with expertise in the field. Disagreements, if any arose, were resolved by consensus. The reader is advised and encouraged throughout the text to refer to the 2002 statement for the scientific basis and classical methodological approach of respiratory muscle function.

Of note, this statement contains additional information on respiratory muscle evaluation in two particular settings, of paediatrics and the ICU; due to manuscript constraints, these two settings are confined quasi-exclusively to the supplementary material, along with more technical and methodological details concerning each section of the article.

Section 1. Respiratory muscle function

1.1. Airway opening, oesophageal and gastric pressures: technical considerations

1.1.1. Pressure measurement

Respiratory muscles have two distinct functions: force development (pressure changes) and shortening (lung volume changes). Several key points must be considered [1]:

- 1) Pressures reflect barometric pressure difference.
- 2) In unaltered physiology/anatomy, specific pressures represent entire corresponding spaces. Gravity/shear-stress affects pressure readings [2]. Figure 1 indicates pressure recording sites.
- 3) Pressure differences are assessed across corresponding structures. Table 1 lists thoracic pressure readings.
- 4) Pressure differences between two points reflect difference across at least two (group of) structures (e.g. chest wall/pleural cavity).
- 5) Pressure measurement reflects global muscle “output” (rather than contractile property *per se*).
- 6) Assessment occurs *via* voluntary manoeuvres or *via* evoked contractions (see below).

1.1.2. Pressure assessment devices

1.1.2.1. Pressure transducers

Frequency response flat up to 10–15 Hz assesses dynamic/static pressures [1]. Transducers should be calibrated in specific settings, since attached systems (e.g. catheters) alter frequency responses [3]. One should ensure identical frequency responses on both sides (differential transducers) [1]. Digital calibration is acceptable; however, a check *via* water manometer should be done regularly [1]. Pressure range should be ± 300 cmH₂O and resolution ≤ 0.5 cmH₂O [1].

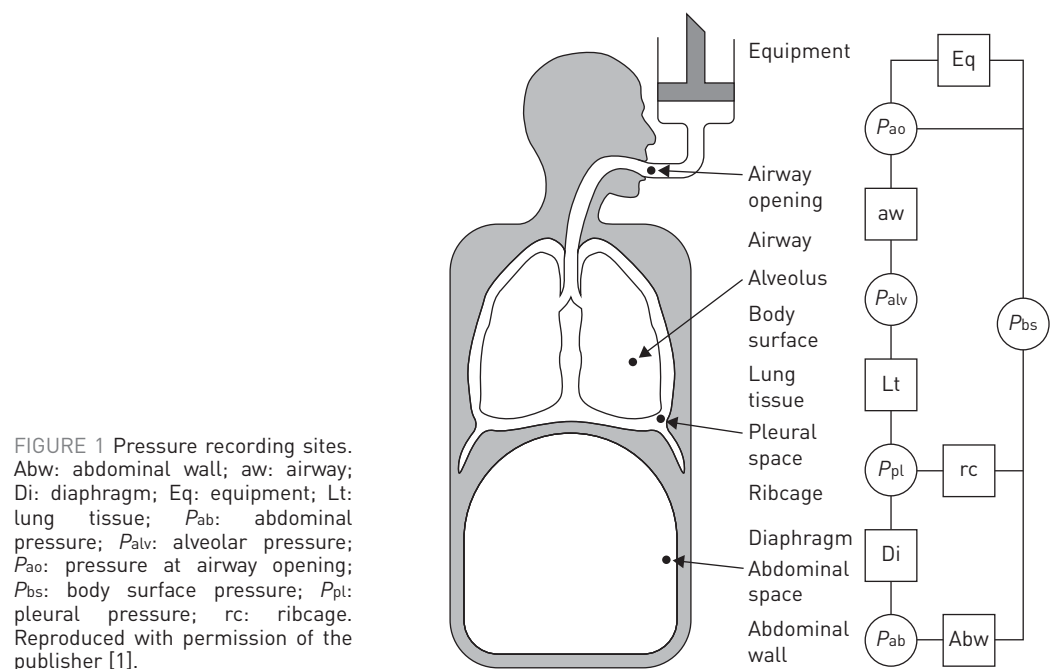


TABLE 1 Thoracic pressure readings

Pressures at a location P_{ao} (airway opening pressure) P_{alv} (alveolar pressure) P_{pl} (pleural pressure) P_{ab} (abdominal pressure) P_{bs} (body surface pressure)**Pressure differences across structures** $P_{el(L)}$ (elastic recoil pressure of the lung (pressure across lung tissue)) P_L (transpulmonary pressure) P_{rc} (pressure across the rib cage) P_{aw} (flow-resistive pressure in airways) P_{cw} (pressure across the chest wall) P_{di} (transdiaphragmatic pressure) P_{rs} (transrespiratory system pressure) P_{abw} (transabdominal wall pressure) P_{eq} (pressure across the equipment)**Relationship among pressures**

$$\left. \begin{aligned} P_{aw} &= P_{ao} - P_{alv} \\ P_{el(L)} &= P_{alv} - P_{pl} \end{aligned} \right\} = P_L = P_{ao} - P_{pl}$$

$$\left. \begin{aligned} P_{rc} &= P_{pl} - P_{bs} \\ P_{di} &= P_{pl} - P_{ab} \\ P_{abw} &= P_{ab} - P_{bs} \end{aligned} \right\} = P_{cw} = P_{pl} - P_{bs}$$

$$\left. \begin{aligned} P_{rs} &= P_{ao} - P_{bs} = -P_{eq} \end{aligned} \right\}$$

1.1.2.2. Probes for invasive pressure assessment

Air-filled balloon catheters are used to record oesophageal (P_{oes} , ~pleural pressure) and gastric pressure (P_{ga} , ~abdominal pressure) [4]. Specific characteristics need to be considered and standardised preparation is required [1, 5]. Certain catheters additionally allow diaphragmatic electromyography (EMG) [1].

Repeated checking of air filling volumes and entire system volume displacement coefficient guarantees adequate balloon inflation [1, 5].

Appropriate system frequency responses (*e.g.* catheter diameter) are crucial for dynamic manoeuvres with high pressure changing rates (*e.g.* sniffs/twitches) [1]. Important characteristics include reasonable stiffness and several spirally arranged catheter holes at balloon portion, to avoid dampened signals [1, 5].

Liquid-filled catheters and catheter-mounted microtransducers have drawbacks (*e.g.* damped pressure signal in oesophagus/stomach or wide limits of agreement) and are not used in this setting [1, 6].

1.1.2.3. Devices for measurement of airway opening pressure

Airway opening pressure (P_{ao}) is usually sampled from side taps ("lateral pressure") located in the mouthpiece/tracheal tube/facemask/nostril plug [1, 7]. Nasal pressure reflects airway pressure only during undisturbed communication between nostrils/mouth with nasal flows [1]. The device to which the side tap is connected must have a cross-sectional area large enough to minimise the Bernoulli effect [8].

For P_{ao} to estimate alveolar pressure during dynamic respiratory efforts against an occluded airway, alveolar-oral pressure transmission must be fast [1]. The transmission time constant depends on airway resistance and compliance of extrathoracic airways (*i.e.* mouth/cheeks/equipment) [1]. This is especially important when airway resistance increases (*e.g.* asthma, chronic obstructive pulmonary disease (COPD)) [1].

1.2. Voluntary tests of respiratory muscle strength**1.2.1. Maximal static inspiratory and expiratory mouth pressure**

Measurements of maximum static inspiratory ($P_{I_{max}}$) or expiratory ($P_{E_{max}}$) pressures at the mouth allow a simple assessment of global respiratory muscle strength in a clinical setting [1]. Tests are volitional and require full subject cooperation. $P_{I_{max}}$ is usually measured at residual volume and $P_{E_{max}}$ at total lung capacity (TLC) to record the maximum value of three manoeuvres that vary by less than 10% (more details can be found in the supplementary material). Measuring $P_{I_{max}}$ at functional residual capacity (FRC) has the advantage of representing the maximal static inspiratory pressure measured at the lung volume at which patients breathe tidally; however, it is greatly influenced by the level of lung hyperinflation or the severity of restriction, so careful attention should be paid under these conditions.

P_{Imax} is strongly related to exertional dyspnoea (figure S4) [9]. The test might also serve as a screening instrument to identify patients with respiratory muscle weakness (figure 2, and supplementary material) [10]. Results should not be interpreted in isolation but together with the overall clinical picture (pathology, symptoms, and load/capacity balance during daily activities). The test is responsive to evaluate changes within subjects. Characteristics of studies that provide reference values for P_{Imax} and P_{Emax} measurements are summarised in the supplementary tables S2–S8 [11]. Measurements of mouth pressures are also used in cooperative children older than 6–8 years of age (table S14), and to evaluate muscle strength in the ICU (supplementary material).

1.2.2. Maximal sniff nasal inspiratory pressure

During measurement of maximal sniff nasal inspiratory pressure (SNIP), inspiratory pressure is recorded by a pressure transducer connected to a catheter placed in the nostril [12]. The test is performed at FRC. The subject is instructed to sniff quickly and deeply. SNIP has been validated in healthy individuals [12] and patients with COPD [13], and is also very useful for children >2 years of age [14]. Precision is good in healthy subjects without severe nasal congestion. Even in COPD there is good repeatability [13]. More information including normative values is presented in supplementary table S9.

1.2.3. Peak cough flow

Peak cough flow (PCF) estimates the effectiveness of mucus clearance and expiratory muscle function in neuromuscular disorders [15, 16]. The measurement is performed with subjects seated. An oronasal mask/mouthpiece is connected to a pneumotachograph or peak flow meter. Subjects are instructed to perform a maximal cough after complete inhalation [17]. They should perform 3–6 manoeuvres (<5% variability) and the maximum PCF ($\text{L}\cdot\text{min}^{-1}$) should be reported [17]. In NMDs, (manually) assisted PCF might be appropriate [18]. Hand-held peak flow meter devices might overestimate PCF if spirometer recordings of PCF are $<270 \text{ L}\cdot\text{min}^{-1}$ [17].

PCF informs the need to start manual/mechanical exsufflator/insufflator therapy because PCF $<270 \text{ L}\cdot\text{min}^{-1}$ is associated with higher likelihood of pulmonary complications in neuromuscular disorders [17]. Healthy (children) adults achieve PCF measurements of approximately (150) 470–600 $\text{L}\cdot\text{min}^{-1}$; PCF

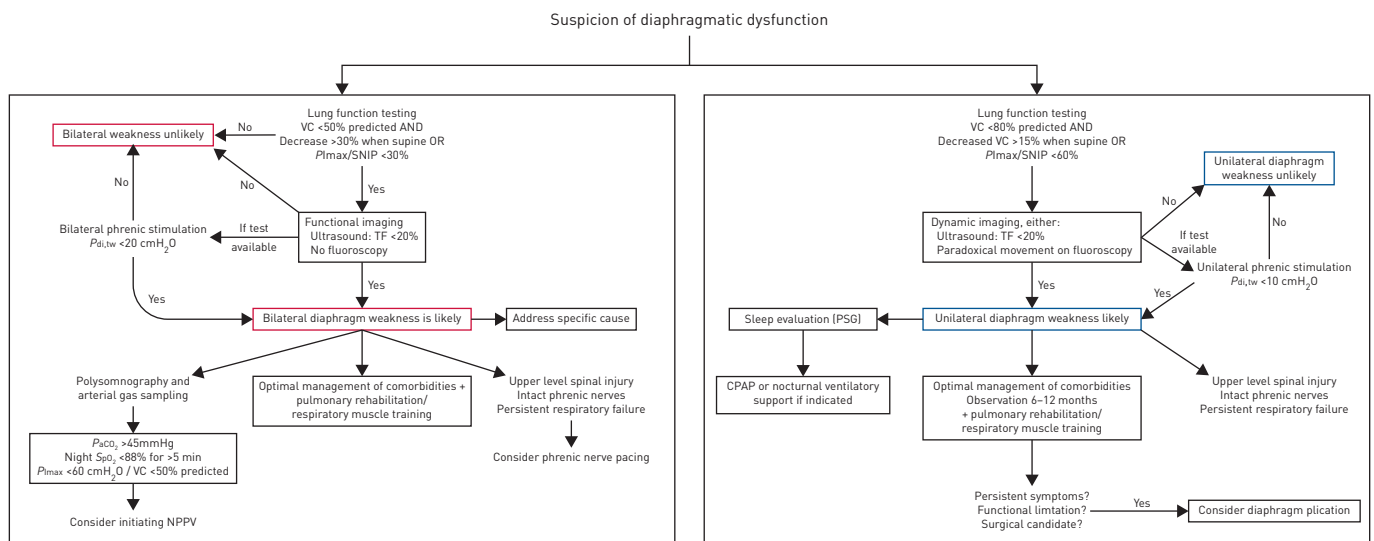


FIGURE 2 Expert opinion on the suspicion of diaphragmatic dysfunction. The figure describes the current practice of how the members of the task force suspect and treat respiratory muscle dysfunction (especially for unilateral and bilateral diaphragm weakness), outside of the intensive care setting (this is, however, not intended as a recommendation for clinical practice). In the absence of clearly defined lower limits of normal, it has long been accepted that a P_{Imax} or sniff- P_{di} or $P_{\text{dimax}} \geq 80 \text{ cmH}_2\text{O}$ in men and $\geq 70 \text{ cmH}_2\text{O}$ in women, and/or $\text{SNIP} \geq 70 \text{ cmH}_2\text{O}$ in men and $\geq 60 \text{ cmH}_2\text{O}$ in women are generally thought to exclude clinically significant inspiratory muscle weakness [1], and unilateral and bilateral diaphragm paralysis can be expected to decrease P_{Imax} or SNIP in the ranges of 60% [41] and <30% [42] of the predicted values, respectively. However, these values may be greatly impacted by the presence of underlying obstructive or restrictive lung disease [40]. A $P_{\text{di,tw}} > 10 \text{ cmH}_2\text{O}$ with unilateral phrenic nerve stimulation or $> 20 \text{ cmH}_2\text{O}$ with bilateral phrenic nerve stimulation also rules out clinically significant weakness [1]. Please refer to the text for more details. SNIP: sniff nasal inspiratory pressure; VC: vital capacity; P_{Imax} : maximal inspiratory pressure; TF: thickening fraction of the diaphragm; PSG: polysomnography; CPAP: continuous positive airway pressure; P_{di} : transdiaphragmatic pressure; $P_{\text{di,tw}}$: twitch transdiaphragmatic pressure; NPPV: noninvasive positive pressure ventilation; PaCO_2 : arterial partial pressure of carbon dioxide; SpO_2 : peripheral oxygen saturation.

$<160 \text{ L}\cdot\text{min}^{-1}$ is associated with higher likelihood of extubation/weaning failure in neuromuscular disorders (more details can be found in the supplementary material) [17, 19].

1.3. Voluntary manoeuvres with oesophageal and gastric pressures

Measurements of P_{oes} , P_{ga} and transdiaphragmatic pressure ($P_{\text{di}}=P_{\text{ga}}-P_{\text{oes}}$) while sniffing and coughing are useful when noninvasive measures of in- and expiratory muscle function (e.g. SNIP or PCF) provide equivocal information. Assessments during sniff are particularly useful when SNIP yields suspiciously low values, e.g. in patients with upper airway obstruction (hypertrophy of the adenoids, rhinitis, polyps) or lower airway obstruction. Assessments during cough are needed to assess the expiratory muscles when glottis function is compromised, for example in patients with bulbar amyotrophic lateral sclerosis (ALS). These measurements may also be used to refine clinical diagnosis [20, 21]. Maximal muscle relaxation rate (MRR) can provide additional information on respiratory muscle function [22, 23] but its clinical application is limited. Measurements of P_{oes} and P_{ga} during voluntary manoeuvres can also be obtained in the ICU and in children (supplementary material).

In adults, the average within-subject, between-occasion coefficient of variation (CV) is 11% for sniff- P_{di} [24] and 6.9% for cough- P_{ga} (table 2) [21]. No such values are available for children. For sniff-MRR, within-subject and between-occasion CVs range from 6 to 26% [25].

Reference values are given in table 2 where available.

In many diseases, pressures produced during sniff and cough are less than normal, both in adult patients (e.g. heart failure [26], stroke [27], COPD [25], pulmonary fibrosis [25], cystic fibrosis [28] and NMDs [20, 21, 29]) and in children with NMDs [30, 31]. In a cohort of patients with mixed diagnoses [20], adding SNIP to P_{Imax} reduced the false-positive diagnosis of inspiratory muscle weakness by 20% (with sniff- P_{di} not adding more diagnosis accuracy). Adding cough- P_{ga} to P_{Emax} decreased false-positive diagnosis of expiratory muscle weakness by 30% [20].

The use of sniff- P_{di} and cough- P_{ga} has not been widely explored for prognosis. In ALS patients, sniff- P_{di} correlated with SNIP [32] and SNIP $<40 \text{ cmH}_2\text{O}$ was associated with desaturation during sleep; hazard ratio for death was 9.1. Sniff- P_{di} , sniff- P_{oes} and twitch P_{di} ($P_{\text{di,tw}}$, see below) were significant predictors of ventilation-free survival in ALS patients [33], while P_{Emax} and transdiaphragmatic pressure elicited by phrenic nerve stimulation ($P_{\text{di,tw}}$) were predictors of absolute survival.

After lung volume reduction surgery [34], sniff- P_{di} , SNIP and P_{Imax} increased significantly, while 8 weeks of rehabilitation did not add any further improvement [34].

In COPD, after exhaustive treadmill walking, sniff- P_{oes} did not change significantly; sniff- P_{oes} -MRR decreased by 42%, and recovered within 5 min of rest [35].

1.4. Respiratory muscle-related mechanics of breathing

1.4.1. Lung function testing

Pulmonary function tests, especially measurements of upright and supine vital capacity (VC), which depends on activation of both inspiratory and expiratory muscles [36], are noninvasive and readily available measurements contributing to the evaluation of respiratory muscle function, especially the diaphragm [36–39]. Unilateral diaphragm weakness is usually associated with a modest decrease in VC, to approximately 75% of predicted [40, 41], with a further 10–20% decrease in the supine position (15% which represents twice the CV of the measure could be considered the lower limit of normal) (figure 2) [41], while FRC and TLC are usually preserved [40, 41]. In severe bilateral diaphragm weakness, VC is usually 50% of predicted and can further decrease by 30% or more when supine [42]. A normal supine VC makes the presence of clinically significant diaphragmatic weakness unlikely.

TLC can also be reduced (70–79% of the predicted value with mild weakness, up to 30–50% of the predicted value in moderate-to-severe weakness), while residual volume can be elevated [43]. Of note, in patients with diaphragm weakness, the magnitude of fall in VC in the supine position correlates with the reduction in sniff- P_{di} [43].

In many NMDs [44–50], such as ALS, a significant reduction of VC at diagnosis, as well as its rate of decline over time, are recognised as criteria for initiating noninvasive ventilation [51, 52]. Reduction in VC is also predictive of sleep disordered breathing, respiratory failure, worse prognosis and response to treatment, to a lesser extent, with good sensitivity (80–95%) but quite variable specificity (50–90%) [53].

1.4.2. Indices of respiratory muscle effort

The pressure output of the respiratory muscles can be assessed by calculating 1) the work of breathing (WOB), 2) the pressure–time product (PTP) of either the oesophageal pressure (PTP $_{\text{oes}}$; reflecting

TABLE 2 Characteristics of the main voluntary and evoked manoeuvres to assess respiratory muscle strength

Tests	Main variables	Reference values and discriminative values	Repeatability/reliability/validity	Cautions	Setting (expert centres, general clinical use, research, etc.)	Remarks
Voluntary manoeuvres with mouth pressure	P_{Imax}	Yes (tables S2–S4)	Sufficiently repeatable and reliable measurements in untrained subjects (<10% variability between efforts) can usually be obtained within 5 efforts [297]. Peak values are typically reached after 9 attempts [298].	Standardisation of lung volumes, mouthpiece and recorded pressure (peak <i>versus</i> plateau) required.	SNIP and mouth pressures can be used in clinical practice after thorough training of the procedures.	Always to be interpreted in clinical context of symptoms and diagnosis.
	P_{Emax}	Yes (tables S5 and S6)	Reliable peak values usually achieved after 5–6 efforts. Within subject between occasion coefficient of variation around 10% [21].	Standardisation of lung volumes, mouthpiece and recorded pressure (peak <i>versus</i> plateau) required.		Always to be interpreted in clinical context of symptoms and diagnosis.
	SNIP	Yes (table S9)	Yes. Possibly fewer efforts needed for acceptably reliable measurements in comparison to P_{Imax} in untrained subjects [12, 13, 20, 299, 300].	Cautions in subjects with severe nasal congestion. Although SNIP and P_{Imax} has a good correlation, the agreement between these two methods is variable. Thus, they are complementary and not interchangeable in the evaluation of inspiratory weakness	SNIP in association with P_{Imax} reduces the false-positive diagnosis of inspiratory weakness by nearly 20% [5].	Should be used as a complementary variable (<i>i.e.</i> in addition to a first screening with mouth pressures) to investigate inspiratory weakness. Always use the reference values of your population when available.
	PCF	Healthy subjects: 468–588 L·min ⁻¹ [301] Increased extubation/weaning failure <160 L·min ⁻¹ in NMD patients [302]	No sufficient data available. Be careful with dose of local anaesthesia.	At least 3–6 PCF with <5% variability need to be assessed [17]	Simple to assess. Especially useful in NMD patients.	No direct link between “cut-off” values and clinical consequences (<i>e.g.</i> cough assist).
Voluntary manoeuvres with oesophageal and gastric pressures	Sniff	No normal values exist; mean±SD (range) achieved by healthy subjects: P_{di} [37 M]: 148±24 [111–124, 126–204] cmH ₂ O [303] P_{di} [27 F]: 122±25 [82–182] cmH ₂ O [303] P_{di} [64]: 136±37 [82–204] cmH ₂ O [303] P_{di} [32]: 134±24 [86–195] cmH ₂ O [24] P_{oes} [37 M]: 105±26 [52–150] cmH ₂ O [303] P_{oes} [27 F]: 92±22 [52–140] cmH ₂ O [303]	CV- P_{di} (healthy adults): 11% [24]	NA	Expert centre research	SNIP/sniff- P_{oes} (children): CF 0.72±0.13 [28] NMD patients 0.83±0.17 [28] Thoracic scoliosis 0.86±0.10 [28] 3-year ventilator-free survival in ALS patients: sniff- P_{di} cut-off 108.5 cmH ₂ O (sensitivity 0.85, specificity 0.98) [33]

Continued

TABLE 2 Continued

Tests	Main variables	Reference values and discriminative values	Repeatability/reliability/validity	Cautions	Setting (expert centres, general clinical use, research, etc.)	Remarks
		P_{oes} [64]: 100±25 [52–150] cmH ₂ O [303] P_{oes} [12]: 93±27 cmH ₂ O [28] P_{ga} [37 M]: 43±32 [0–134] cmH ₂ O [303] P_{ga} [27 F]: 29±29 [0–108] cmH ₂ O [303] P_{ga} [64]: 37±31 [0–134] cmH ₂ O [303]				
	Cough	Normal values [21]: P_{ga} [62 M]: 214±42 cmH ₂ O P_{ga} [37 F]: 165±35 cmH ₂ O Lower limits of normal [21]: 132 cmH ₂ O [62 M], 97 cmH ₂ O [37 F]	CV- P_{ga} (healthy adults): 6.9% [21]	NA	Expert centre research	Cough- P_{ga} assessment is helpful for patients with low P_{Emax} to avoid false-positive diagnosis of expiratory muscle weakness.
Evoked manoeuvres	$P_{mo,tw}$	Possible diaphragm weakness $P_{mo,tw}$ <–11 cmH ₂ O (cervical magnetic stimulation) $P_{mo,tw}$ <–8 cmH ₂ O (bilateral electrical stimulation)				
	$P_{di,tw}$	Possible diaphragm weakness $P_{di,tw}$ <15 cmH ₂ O				

P_{lmax} : maximal inspiratory pressure; P_{Emax} : maximal expiratory pressure; SNIP: maximal sniff nasal inspiratory pressure; PCF: peak cough flow; NMD: neuromuscular disease; P_{oes} : oesophageal pressure; P_{ga} : gastric pressure; P_{di} : transdiaphragmatic pressure; P_{mo} : mouth pressure; $P_{di,tw}$: twitch transdiaphragmatic pressure; $P_{mo,tw}$: twitch mouth pressure; ALS: amyotrophic lateral sclerosis; CF: cystic fibrosis; M: male; F: female; CV: coefficient of variation; NA: not applicable.

the effort done by all of the respiratory muscles), the transdiaphragmatic pressure (PTP_{di} ; reflecting mostly the effort done by the diaphragm) [54], 3) the tension-time index ($TTI = P_t / P_{tmax} \times t_i / t_{tot}$) or 4) the P_{oes} swing. Details on how these indices are calculated, their physiological underpinning, advantages and disadvantages are described in the supplementary material. PTP analyses have been used as an alternative to WOB to quantify respiratory muscle effort in both healthy subjects [55, 56] and patients with COPD [57–59]. PTP is more closely related to respiratory muscle oxygen consumption than WOB [60]. The average value for PTP in healthy subjects is around $100 \text{ cmH}_2\text{O} \cdot \text{s} \cdot \text{min}^{-1}$ while in acute respiratory failure, the average PTP has been reported to be about four-fold greater [61]. During trials of weaning from mechanical ventilation, PTP_{oes} can predict weaning failure [61]. PTP is greater in COPD patients during exercise compared with age- and sex-matched healthy controls [62].

In the clinical setting, inspiratory effort can be simply monitored by measuring tidal swings in P_{oes} ($P_{oes,tid}$) (figure 3) [63]. $P_{oes,tid}$ can serve as an index of global respiratory muscle effort during exercise in patients with chronic respiratory diseases [64]. $P_{oes,tid}$ can identify differences in disease severity between patients with COPD (*i.e.* by Global Initiative for Chronic Obstructive Lung Disease stages) [65] and it is sensitive to changes over time and to interventions [65]. Increases in $P_{oes,tid}$ relative to stable tidal volume responses are related to the perception of dyspnoea during exercise [64, 66]. $P_{oes,tid}$ has been successfully applied as a bedside monitoring tool in sleep studies [67], and during weaning trials [68]. $P_{oes,tid}$ (in analogy with the PTP_{oes}) showed larger changes over the course of a failed weaning trial than breathing pattern parameters (rapid shallow breathing index) [61, 68].

Reduction of resistive and elastic load by continuous positive airway pressure or inspiratory pressure support can reduce inspiratory effort. During exercise, these reductions in inspiratory effort decrease exercise-associated dyspnoea and improve exercise tolerance in patients with COPD [57, 69]. Similar results can be obtained using helium hyperoxia and bronchodilators [70]. It is difficult to establish a minimal clinically important difference of indices of respiratory muscle effort, given the paucity and heterogeneity of the studies. Nonetheless, a clinically meaningful difference of 14–16% from baseline condition has been shown to correlate with a clinically meaningful reduction of exertional dyspnoea after pharmacological intervention such as bronchodilators for both PTP and $P_{oes,tid}$ [71–73]. Finally, exercises that promote slow and deep breathing have the potential to reduce the elastic component of WOB and thereby reduce inspiratory effort [74–76]. In addition, changes in inspiratory duty cycle (decreased t_i / t_{tot}) induced by these breathing techniques can further reduce PTP by reducing inspiratory time per minute [77]. Measurements of pressure during brief inspiratory occlusions (typically 0.1 s) applied without warning before the individual recognises the occlusion and reacts (*i.e.* $P_{0.1}$) can be a useful index of respiratory centre motor output (supplementary material).

1.5. Evoked manoeuvres

Non-volitional evaluation of diaphragm (dys)function and fatigue (*i.e.* a reduction in the ability to produce force/pressure following contractile activity) can be performed by phrenic nerve stimulation; diaphragm

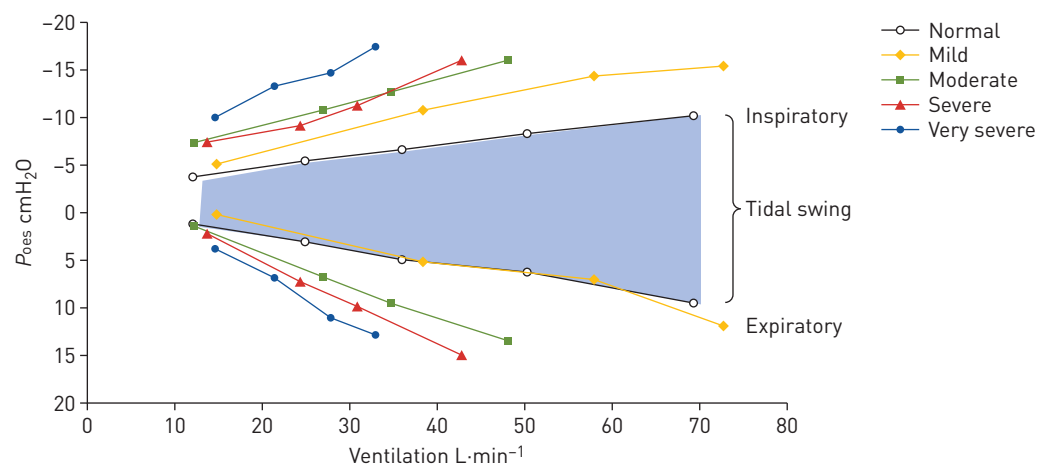


FIGURE 3 Tidal oesophageal pressure (P_{oes}) swings are shown with varying severity of chronic obstructive pulmonary disease and in age-matched healthy control subjects. As disease severity worsens, the amplitude of inspiratory and expiratory P_{oes} increases for a given ventilation during exercise. The shaded area represents the tidal P_{oes} swing in the healthy control subjects. Original data from the authors' laboratory. Values are means.

contraction causes a sudden and short-lasting fall in P_{oes} and a rise in P_{ga} , the difference providing $P_{di,tw}$. Abdominal muscles can be evaluated by stimulation of thoracic nerve roots with measurement of gastric pressure ($P_{ga,tw}$).

Magnetic phrenic nerve stimulation has superseded electrical stimulation, except where patients have pacemakers or other implanted electronic devices where magnetic stimulation is contraindicated. Technical considerations during phrenic nerve stimulation include: 1) stimulation must be supramaximal, 2) potentiation resulting from prior contraction must be avoided or standardised, 3) lung volume must be standardised, and 4) different magnetic stimulation techniques (e.g. cervical, bilateral anterior) yield different results and should be used consistently. A noninvasive estimate of $P_{di,tw}$ can be obtained by measuring pressure changes in the upper airway or the mouth ($P_{mo,tw}$) [78] although oesophageal twitch pressure ($P_{oes,tw}$) and thus $P_{mo,tw}$ are more influenced by lung volume than $P_{di,tw}$.

Resting values of $P_{ga,tw}$ have a slightly higher variability (CV 9–10%) than $P_{di,tw}$ (6%).

Age- and sex-specific normal values for adults are lacking, but a cut-off for $P_{di,tw}$ of 18 cmH₂O has been suggested for diagnosis of diaphragm weakness [20]. In healthy subjects, the mean between-occasion variability in $P_{di,tw}$ is $20 \pm 11\%$ and CV 11% [79]. The limit of agreement of the difference of $P_{di,tw}$ recordings is ± 6 cmH₂O [79]. Variations in $P_{di,tw}$ with disease are shown in table 3.

Exhaustive exercise in healthy subjects can elicit a fall in $P_{di,tw}$ (showing diaphragm fatigue), while in a variety of disease states exercise-induced diaphragm fatigue has been reported by some but not all investigators (please refer to supplementary material for more details). Of note, diaphragm fatigue is not necessarily related to exercise performance and development of fatigue may not predict clinical outcomes [80].

Normal $P_{di,tw}$ values are available for neonates [81], infants [82] and children [83], in whom stimulation is acceptable with local anaesthesia. In critically ill patients, measurement of $P_{ao,tw}$ and $P_{di,tw}$ can be particularly useful and several large case series yielded $P_{ao,tw}$ or $P_{di,tw}$ values [84, 85] that are lower than those recorded in healthy individuals (please refer to supplementary material for further details).

1.6. Respiratory muscle endurance testing

Different approaches can be used to assess respiratory muscle endurance: 1) incremental load testing, 2) constant load testing and 3) time trials. Different tests were developed within these categories: 1) a stepwise load increase by increasing resistance/threshold load or minute ventilation, 2) sustaining a given resistance/threshold load or hyperpnoea level to task failure and 3) time trial, where a maximum ventilation (with/without additional resistance) must be achieved within a given duration. While resistive/threshold loading tests mostly apply inspiratory loads [86], hyperpnoea tests load both inspiratory and expiratory muscles [87].

Since test performance is influenced by breathing pattern, breathing frequency and tidal volume should be controlled (feedback) and/or reported [88–90]. When testing pre/post-interventions, starting load/ventilation and increments (if present) need to be identical.

TABLE 3 Summary of the main causes of perturbation in twitch transdiaphragmatic pressure ($P_{di,tw}$)

$P_{di,tw}$ observation	Partitioning	Interpretation	Consider
$P_{di,tw} \uparrow$	$P_{oes,tw} \uparrow$, $P_{ga,tw} \uparrow$	- Strong patient - Potentiased muscles	
$P_{di,tw} \downarrow$	$P_{oes,tw} \downarrow$, $P_{ga,tw} \downarrow$	- True weakness - Submaximal stimulation (e.g. obesity) - Medical comorbidities	- Neurological exam - Is P_{lmax}/SNIP strong? (supports if so) - Age [304], heart failure [305], pulmonary hypertension [306]
$P_{di,tw} \downarrow$	$P_{oes,tw} \downarrow$, $P_{ga,tw} \leftrightarrow$	Hyperinflation	- Review technique - Investigate for COPD - What is end-expiratory oesophageal pressure? (may reveal intrinsic PEEP) - Check air (in balloon catheter systems)

$P_{oes,tw}$: oesophageal twitch pressure; $P_{ga,tw}$: gastric twitch pressure; P_{lmax} : maximal inspiratory pressure; SNIP: sniff nasal inspiratory pressure; COPD: chronic obstructive pulmonary diseases; PEEP: positive end-expiratory pressure.

1.6.1. Maximal incremental load testing

Resistive/threshold testing requires subjects to breathe against a resistive/threshold [1, 89] or tapered flow-resistive [86] load that is increased at regular intervals (minutes or number of breaths), *e.g.* by 10% of baseline P_{limax} until task failure. Inspiratory muscle endurance can be defined as the pressure of the last completed step.

Hyperpnoea testing uses stepwise increasing minute ventilation (*e.g.* +8% of maximal voluntary ventilation (MVV) every 3 min) [1]. It needs special equipment to assure normocapnia and has been increasingly applied in recent years [90]. Ventilatory levels achieved in this test were found to be similar to levels reached in the traditional maximal sustainable ventilation test [91, 92]. Normal values have been established for healthy subjects [90].

1.6.2. Constant load testing

During resistive/threshold testing, subjects are instructed to breathe against a submaximal load [86, 88, 89] until task failure. It has been proposed that the selected load should result in a time to task failure (t_{lim}) of 5–10 min, such that post-intervention test durations can be limited to about 15–20 min without important ceiling effects [86, 89]. Main outcomes are t_{lim} and/or total external work performed during the test [86]. The pattern of breathing during such a test is important and must be taken into account when analysing the data.

During hyperpnoea testing, subjects breathe at a constant ventilation (40–70% MVV) to achieve task failure within 8–12 min.

1.6.3. Time trial

The 10–15 s MVV manoeuvre is too short-lasting for assessment of respiratory muscle endurance. Different protocols exist for testing maximal sustainable ventilation, *i.e.* the ventilation that can be sustained for a given, extended period of time (*e.g.* 12–15 min). However, there is no consensus on which protocol to use for this kind of test [1].

The attraction of these different respiratory muscle endurance tests is that they provide a method for evaluating global respiratory muscle endurance in a single test session. The tests are noninvasive and relatively well-tolerated. Several studies showed large improvements in respiratory muscle endurance after respiratory muscle training by using these tests (supplementary material).

Section 2. Respiratory muscle neurophysiology

Respiratory muscle neurophysiological testing includes 1) EMG to measure the output of the respiratory motor neurons, 2) electroencephalography (EEG), which tests the involvement of motor and premotor areas, and 3) transcranial magnetic stimulation (TMS) which assesses the neural pathways to the respiratory muscles (figure 4).

2.1. Electromyography

EMG is the technique that quantifies the electrical activity of muscles and is used in research and clinical practice to assess respiratory muscle control at rest and during exercise, including estimation of respiratory motor output (as previously reviewed [93, 94]), neuromechanical coupling during loaded breathing [95] and the efficacy of muscle contraction when coupled with measurements of ventilation [96, 97]. Finally, EMG can also be used in the diagnosis of myopathic and neuropathic diseases [1]. A thorough review of the theoretical background and methodology of respiratory EMG recordings is available [1].

Respiratory EMG can be recorded with surface electrodes, an oesophageal electrode inserted *via* the nose, and intramuscular wire or needle electrodes. Appropriate selection of electrodes depends on the EMG technique (*e.g.* physiological recordings *versus* evoked responses), the target muscle, signal reliability and safety (table 4).

Respiratory EMG measurement is usually contaminated with ECG readings, which should be eliminated [98, 99] or excluded from EMG measures. Moreover, respiratory EMG recordings, especially with surface electrodes, are subject to electromagnetic interference [1, 94, 100], contamination from adjacent muscles and changes in lung volume or posture [1, 101]. Diaphragm EMG can be quantified with a multi-pair oesophageal electrode [102, 103], usually standardised to a maximal value (table 4). Given its noninvasive nature, surface parasternal intercostal EMG has been proposed as an alternative measure of respiratory motor output, respiratory load-capacity balance and, potentially, lung disease severity [104–107], but may not be useful during exercise testing [108]. The single motor unit technique can accurately assess respiratory motor output [93] and avoids many of the caveats related to contamination of EMG signals

and normalisation. For evoked responses, normal values of phrenic nerve conduction time are available using either electrical or magnetic stimulation (table 4) [1, 94, 109, 110].

The reliability of these respiratory EMG techniques is reported in table 4.

Respiratory EMG has been used to assess respiratory muscle control in cardiorespiratory disease at rest (table S10) and during exercise (table S11). Briefly, diaphragm EMG is a surrogate of respiratory effort [102, 111–113], it can be used to distinguish between central and obstructive sleep apnoea events [111, 112, 114], to assess exertional breathlessness during exercise [64, 97, 115, 116] and, when combined with VT recordings, it can be used to assess upper airway resistance [112, 117]. Given increased respiratory motor output to the respiratory muscles in COPD [118, 119] and the relationship between EMG and lung function, respiratory EMG has been taken as a marker for disease severity in stable COPD [103] and to predict COPD exacerbations [105], early hospital admission [107] and the effect of medical interventions [120–122].

In the ICU, recordings of the electrical activity of the crural diaphragm (EAdi) using a dedicated nasogastric tube with EMG electrodes has greatly facilitated bedside monitoring of diaphragm activity in both paediatric [123] and adult patients [124]. The EAdi signal can be used to trigger and determine the level of assistance during mechanical ventilation, *i.e.* “neutrally adjusted ventilatory assistance” [125]. The ratio of actual EAdi to maximum EAdi can be used to estimate the patient’s effort to breathe [126]. EAdi is a promising tool for diaphragm activity monitoring, especially during the weaning phase [127].

2.2. Electroencephalography

Respiratory-related cortical networks are not normally activated during resting breathing [128], carbon dioxide stimulation [128], or the ventilatory response to exercise [129]. In contrast, these networks are engaged during voluntary respiratory manoeuvres (apnoea, sniffing or hyperventilation) [128, 130, 131]. They are also engaged when the respiratory system is used for non-respiratory purposes, such as speech [132]. Induction of respiratory neuroplasticity using repetitive TMS have suggested these networks exert a tonic excitatory influence on breathing during wakefulness [133]. The respiratory-related

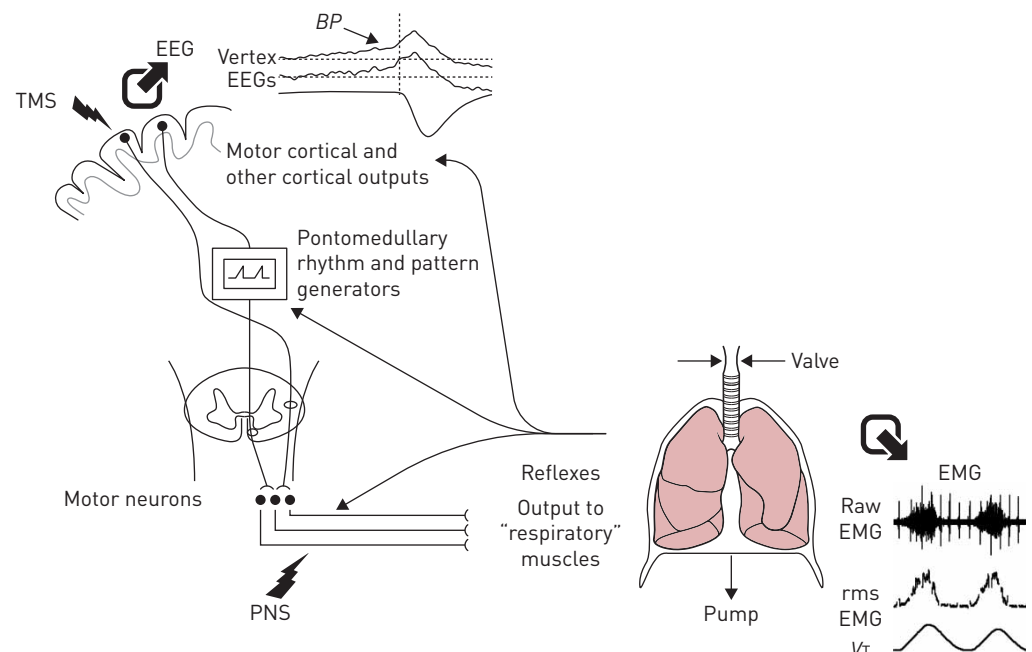


FIGURE 4 Neurophysiological techniques to assess respiratory muscle control. Schematic of the neural control of the human respiratory muscles. Multiple descending pathways integrate at the respiratory motor neurons (with reflex afferent inputs) and determine the functional output of the muscles. Using electromyography (EMG), the output can be measured during resting breathing, exercise and voluntary manoeuvres or as evoked signals in response to transcranial magnetic stimulation (TMS) over the motor areas or phrenic nerve stimulation (PNS) of the peripheral nerve. The output from cortical networks can be measured using electroencephalography (EEG) as the presence of a *bereitschaft* (readiness) potential (BP) indicates respiratory-related cortical activity. rms: root mean square; VT: tidal volume.

TABLE 4 Characteristics of electromyography (EMG) techniques at rest

Tests (EMG techniques)	Main variables	Reference values and discriminative values	Repeatability/reliability/ validity	Cautions/safety	Setting (clinical, research)	Remarks
EMG during breathing. For surface and oesophageal recordings, raw EMG or integral/root mean square is typically normalised to maximal EMG measured during maximal inspiratory efforts (SNIP, $P_{I\max}$, inspiration to TLC and MVV). For the single motor unit technique recorded with needle or wire electrodes, the peak discharge rate is typically reported.	sEMGpara, EMGpara%max	Reference values for men and women, with and without a mouthpiece, raw <i>versus</i> normalised signal [106].	Negligible bias for raw and normalised EMG between recording sessions. Small bias in raw EMGpara for repeat measures in the same recording session [106].	Considered safe except for small chance of skin abrasion during electrode preparation. However, signal is subject to contamination from other muscle activity and movement of muscle.	Clinical, research	Recordings of sEMGpara show promise as a noninvasive method to measure neural respiratory drive [104].
	sEMGscal	NA	Ensemble average of 80 breaths had comparable timing of inspiratory activity as iEMG recordings for 3 participants [307].		Research	sEMGscal has been proposed as a monitoring tool in the ICU [100].
	sEMGdi	NA for adults. Reference values for children during sleep [308].	Excellent reliability within participants, and excellent agreement between occasions and between observers, but data from children who were snorers [308].		Clinical, research	Surface EMG over the chest wall can be very susceptible to contamination.
	oesEMGdi%max	Reference values for young (<50 years) and old (>50 years) subjects. No difference if signal normalised to max in voluntary manoeuvres or evoked response (<i>i.e.</i> oesCMAPdi) [103]	Good repeatability between recording sessions and between observers [103].	Not for use in patients with oesophageal varices.	Clinical, research. Used in neutrally adjusted ventilatory assistance.	The preferred technique for testing respiratory muscle control during exercise given its specificity and safety advantages. Disadvantage of the normalisation procedure is that "maximal efforts" can be, in fact, submaximal [309].
	iEMGdi	No data available for amplitude during quiet breathing. This measure is typically used to compare activity between experimental procedures [310].		Usual considerations with needle insertion (bleeding, pain and infection). Risk of pneumothorax can be minimised with appropriate precautions (<i>e.g.</i> ultrasound and online audio/visual feedback), but greater risk during exercise due to increased lung excursion and chest wall movement.	Research	Even intramuscular recordings are susceptible to cross-talk [311], although to a much smaller degree than surface recordings. Can be used for single- or multi-unit recordings.
	SMUdi, SMUia	Multiple studies in small samples of healthy subjects are available [refer to [93] for references].	Excellent validity given recordings do not need to be normalised, are much less susceptible to recordings artefacts.	Safety considerations as above.	Used in research, and occasionally clinically, in expert centres.	Recorded using needle or selective wire electrodes. A needle electrode can be manipulated in the muscle to sample populations of respiratory motor units.

Continued

TABLE 4 Continued

Tests (EMG techniques)	Main variables	Reference values and discriminative values	Repeatability/reliability/ validity	Cautions/safety	Setting (clinical, research)	Remarks
Evoked signals Measured as the compound muscle action potential in response to electrical stimulation or magnetic stimulation over the cervical spinal cord (CMS) or anterolaterally on the neck (unilateral; UMS) of the phrenic nerve(s).	sCMAPdi	Typically, latency 6–8 ms, depending on stimulation technique or side [109, 312]. Amplitude of CMAP more variable.	Latency is reproducible for both electrical and cervical magnetic stimulation [109].	For magnetic stimulation, the contraindications are listed in the supplementary table.	Both clinical investigation and research.	Signal free of contamination if phrenic nerve is activated without co-stimulation of other muscles. Usually used to diagnose neuromuscular diseases.
	oesCMAPdi	Using a multi-pair electrode, latency is 6–8 ms [102, 313]. Latency shorter on right <i>cf.</i> left side and shorter compared to sCMAPdi from costal diaphragm [313]. Amplitude of CMAP is more variable [313].	Good reproducibility between recording sessions for latency [102, 313]. Good agreement between electrical and unilateral magnetic stimulation for latency and amplitude [102].	Safety considerations for magnetic stimulation as above. Oesophageal catheter not for use in patients with oesophageal varices.	Clinical, research.	

s: surface recordings; ia: intercostal/accessory muscles; oes: oesophageal; para: parasternal intercostal muscle of the second interspace; scal: scalene muscle; di: diaphragm; %max: as a percentage of maximal EMG; SNIP: sniff nasal inspiratory pressure; P_{Imax} : maximal inspiratory pressure; TLC: total lung capacity; MVV: maximal voluntary ventilation; ICU: intensive care unit; SMU: single motor unit; CMS: cervical magnetic stimulation; UMS: unilateral magnetic stimulation; CMAP: compound muscle action potential; NA: not applicable.

cortico-subcortical networks are also engaged in situations where the breathing control system is challenged. Thus, a cortical drive to breathe contributes to the maintenance of ventilatory activity during wakefulness, in spite of profound hypocapnia [134]. Respiratory-related cortico-subcortical networks are also activated when the respiratory system is faced with mechanical constraints [128, 135–138]. This activation is not only sensory, but also motor. A motor respiratory-related cortical activity has been described in various clinical situations. Patients with deficient respiratory automaticity due to Phox 2B mutations (congenital central alveolar hypoventilation) exhibit respiratory-related cortical activity on their electroencephalograms during resting breathing [139]. Detailed observations made in one such patient showed better cognitive performance during mechanical ventilation than during unsupported breathing [140]. This finding lends support of the actual role of the cortical activity in sustaining ventilation (“dual tasking paradigm” [141]). A similar cortical activity has been described in patients with severe forms of the obstructive sleep apnoea syndrome (OSAS) [142] (probably related to the inspiratory load induced by upper airway abnormalities) and in patients with inspiratory muscle weakness due to ALS [143]. Experimental and clinical data are therefore consistent with the notion that the respiratory-related cortical networks provide cortico-medullary co-operation when automatic breathing is compromised. Activation of respiratory-related cortical networks in response to experimental loading is accompanied by respiratory discomfort [137, 138, 143]. In patients with diaphragm dysfunction, alleviating dyspnoea by mechanical ventilatory assistance silences respiratory-related cortical activity [143], suggesting a causative relationship. These observations have led to the hypothesis that respiratory-related EEG activity could constitute a surrogate for self-reported dyspnoea in patients unable to directly communicate with their caregivers, thus forming the basis for a patient–ventilator interface [144]. Of note, the motor cortical activities related to breathing are not synonymous of breathing discomfort (*e.g.* voluntary respiratory manoeuvres), and it must be kept in mind that the brain correlates of breathing discomfort are numerous, very complex and mostly sensory in nature (as exemplified by a host of specific studies that are beyond the scope of this statement).

2.3. Transcranial magnetic stimulation

Transcranial magnetic stimulation (TMS) is a widely used noninvasive neurophysiological technique to assess the excitability of the cerebral cortex and of the corticospinal tract *in vivo* (table 4) [145].

TMS causes no long-term adverse effects in healthy subjects. High frequency (1–50 Hz), high-intensity repetitive TMS (rTMS), however, has the potential to induce epileptic seizures even in healthy individuals [146]. This can be minimised by careful selection of subjects [147] and stimulus threshold, and strict adherence to the available safety guidelines (table 5, table S12) [55].

The validity of TMS critically depends on the appropriate location of EMG electrodes [148] and control of background muscle activity and noise. In the research laboratory, single- and paired-pulse TMS have been used to document and describe the corticospinal pathway to the diaphragm at rest and during different physiological conditions in healthy subjects (supplementary material) [149–151]. In the clinical field, TMS has been used to document the involvement of the respiratory muscles in patients with neurological conditions, such as stroke and multiple sclerosis [152–154] (table S13).

Test–retest reliability of TMS for respiratory muscles are not available. These data for limb muscles are summarised in table 5.

Results mostly from upper airway and diaphragm muscles in response to TMS are documented. Widespread disease-related alteration of corticomotor excitability (as documented by changes in a hand muscle) could also indirectly influence respiratory muscle control and are summarised in the table S13.

In OSAS patients, genioglossus central motor conduction time (CMCT) closely correlates with severity of disease [155]. An increase in cortical-motoneuronal excitability is observed in the genioglossus and diaphragm muscles of awake OSAS patients [155, 156], but not for submental muscles [157, 158]. No plasticity-related changes in genioglossus cortical activity is observed in response to rTMS trains [159, 160].

In stable patients with COPD, intracortical facilitation (ICF) of the diaphragm correlates with inspiratory muscle strength, whereas intracortical inhibition correlates with arterial partial pressure of CO₂ [161]. In COPD, the corticospinal pathway to the diaphragm is more excitable and intracortical facilitation of the diaphragm is markedly attenuated compared to healthy subjects [162].

In the ICU, diaphragm response to TMS in patients with central ventilatory paralysis (*e.g.* cervical spinal cord lesions) predicts the recovery of spontaneous ventilation within 1 year [163]. In patients with stroke, the respiratory system response to TMS represents a simple bedside technique to assess airway clearance and evaluate aspiration risk [164].

For the use of TMS to evaluate interventions, please refer to table S13.

TABLE 5 Characteristics of transcranial magnetic stimulation (TMS) paradigms and related measures

Tests (TMS paradigms)	Main measures	Definition	Physiological significance	Repeatability/reliability/validity	Safety	Setting (clinical, research)
Single-pulse TMS		Noninvasive and painless neurophysiological technique to evaluate the excitability of motor cortical area and the cortical spinal pathways conductivity through the administration of magnetic stimuli over the scalp.			Carries little risk beyond occasional local discomfort at the site of stimulation or a transient headache in susceptible subjects. No change in blood pressure, heart rate, EEG, serum prolactin level, serum cortisol level, or in a variety of memory, cognitive, learning, sensory and motor tests [314].	
	Motor evoked potential (MEP)	Muscular response obtained after a single TMS pulse applied over the contralateral primary motor cortex at appropriate stimulation intensity.	Integrity of the corticospinal tract and excitability of the corticospinal system.	Moderate to good reliability for MEP amplitude of FDI muscle at rest and under active condition; MEP amplitude is more reliable at 120% intensity of stimulation than those obtained at 100% [315].		Research
	MEP latency	Time interval between the application of the TMS pulse on the motor cortex area and MEP onset from the contralateral target muscle; it reflects the conductivity of both the central and peripheral nervous systems, as well as neuromuscular junctions and muscles.				Research
	MEP amplitude	Amplitude of MEP response measured peak-to-peak. It reflects the excitatory state of output cells in the motor cortex, nerve roots and the conduction along the peripheral motor pathway to the muscles.				Research
	Resting motor threshold (RMT)	Lowest TMS intensity able to evoke MEPs in the resting target muscle when single-pulse stimuli are applied to the motor cortex.	Reflects the excitability of a central core of neurons, which arises from the membrane excitability and a balance between inhibitory and excitatory input from local circuits.	Good reliability in FDI for short- and long-term interval [315], also in ADM [316] and APB, EDC, FCR [317].		Research
	Active motor threshold (AMT)	Lowest TMS intensity required to obtain a MEP response during a weak muscle contraction.		Good to excellent short- and long-term reliability in FDI [315].		Research
	Cortical silent period (CSP)	Period of suppression of EMG activity following a twitch suprathreshold TMS stimulus of a target muscle during a sustained voluntary contraction of this muscle.	Cortico (spinal) inhibitory mechanisms, possibly GABA _B mediated (but not only).	Moderate to good reliability in ADM [315] and FDI [317].		Research
	Central motor conduction time (CMCT)	Latency difference between the MEPs induced by TMS and by peripheral (motor root) stimulation.	Reflects the integrity of the cortical-spinal tract, from the upper to the lower motor neurons.			Research
Continued						

TABLE 5 Continued

Tests (TMS paradigms)	Main measures	Definition	Physiological significance	Repeatability/reliability/validity	Safety	Setting (clinical, research)
Paired-pulse TMS		TMS paradigm to study intracortical inhibitory and excitatory phenomena by means of a conditioning subthreshold stimulus preceding a suprathreshold test stimulus applied at different interstimulus interval.				Research
	Intracortical facilitation (ICF)	Paired-pulse TMS measure obtained with long interstimulus interval where the conditioning stimulus is followed by an enhanced response with respect to the test stimulus; it is modulated by multiple neurotransmission pathways.	Expresses the activity of glutamatergic excitatory circuits	Poor reliability in ADM [315].		Research
	Short latency intracortical inhibition (SICI)	Paired-pulse TMS measure obtained with short interstimulus interval where the conditioning stimulus is followed by an inhibition with respect to the test stimulus; it is attributed to an activation of inhibitory neuronal system transmission.	Reflect the activity of GABAergic inhibitory circuits	Good short-term and long-term reliability under resting, not for active conditions [315].		Research
Repetitive TMS (rTMS)	rTMS	Train of TMS pulses of the same intensity applied at a given frequency to a given brain area, that can transiently influence the function of stimulated and connected brain areas, mainly dependent on stimulation frequency.			Even in normal subjects, prolonged, high intensity, rTMS at 10–25 Hz rates can produce partial seizures with or without secondary generalisation [146]. Short inter-train intervals can cause transient degradation in short term verbal memory immediately following rTMS [318].	Research
	Low-frequency rTMS	Trains of variable duration at ≤ 1 Hz stimulation frequency.	Depression of the excitability of the stimulated regions, possibly <i>via</i> LTD.			Research
	High-frequency rTMS	Trains of variable duration at ≥ 1 Hz stimulation frequency.	Increase of the excitability of the stimulated regions, possibly <i>via</i> LTP.			Research
	Theta burst stimulation (TBS)	A form of complex rTMS trains combining different frequencies (<i>i.e.</i> 50 Hz pulse-trains repeated at a rate of 5 Hz) with after-effects on cortical-spinal and cortical-cortical excitability that may reflect changes in synaptic plasticity.	Inhibition when higher than 1 Hz.			Research

EEG: electroencephalography; LTD: long-term depression; LTP: long-term potentiation; ADM: abductor digiti minimus muscle; FDI: first dorsal interosseous; APB: abductor pollicis brevis; EDC: extensor digitorum communis; FCR: flexor carpi radialis muscles.

Section 3. Respiratory muscle imaging

3.1. Ultrasound

Since the publication of the previous ATS/ERS statement [1], numerous studies have reported on the use of ultrasound to assess diaphragm dimensions and activity. With the increasing availability of ultrasound at the bedside, this technique allows a simple, rapid and direct evaluation of the diaphragm that is more sensitive than fluoroscopy for the identification of muscle activity [165].

The most frequently assessed variables using diaphragm ultrasound are: 1) static measurement of end-expiratory diaphragm thickness (T_{di}); 2) dynamic evaluation of the ratio of inspiratory to expiratory diaphragm thicknesses, reported as thickening ratio (TR; inspiratory thickness/expiratory thickness) or thickening fraction (TF; (inspiratory thickness – expiratory thickness)/end-expiratory thickness); and 3) diaphragmatic excursion [166]. Measurements of T_{di} and TF are performed by placing a high-frequency linear probe at the level of the zone of apposition, while diaphragm excursion is measured using a curvilinear probe placed in the subcostal region and recording diaphragm movements in M-mode (figure 5).

3.1.1. Diaphragm thickness

In healthy subjects at rest, intra- and inter-observer reliability of T_{di} are high [167–171] and ultrasound estimates of T_{di} are correlated to direct anatomical measurements [168]. The lower limit of normal for T_{di} has been reported to be 0.15 cm in healthy subjects, with a wide baseline range of values [167]. Similar values have been reported for patients with COPD [172]. It is unclear whether a T_{di} value below this threshold can be used to identify diaphragm dysfunction. T_{di} does not seem to change with age [167] but can be influenced by posture [173], stature [171, 174] and body composition [171, 175]. In addition, in studies of patients with diaphragm weakness, a large proportion of subjects had T_{di} values of 0.15 cm [176–178]. However, the temporal evolution of T_{di} in these patients was related to the change in VC in those with recovery of diaphragm function [176]. This observation suggests that T_{di} can be used to monitor the evolution of diaphragm weakness [176]. In mechanically ventilated patients, T_{di} is reproducible [179, 180]. T_{di} is not correlated with $P_{ao,tw}$ when patients are receiving assist control ventilation or pressure support ventilation [181]. T_{di} is a poor predictor of weaning outcome [182–184]. Finally, over the course of mechanical ventilation, T_{di} can decrease, increase or remain unchanged [185, 186].

3.1.2. Diaphragm thickening fraction and ratio

The measurement of TF is reproducible [179], with a reported lower limit of normal of 20% in healthy subjects and patients with COPD [167, 172]. A TF around 20%, however, is possibly more closely associated with near complete paresis rather than partial dysfunction, as the mean values for TF in healthy subjects can frequently exceed 100% [167].

Diaphragmatic contractions produce both muscle shortening and thickening. The correlation between diaphragm thickening and diaphragm effort, however, is tenuous: ultrasound measurements of diaphragm

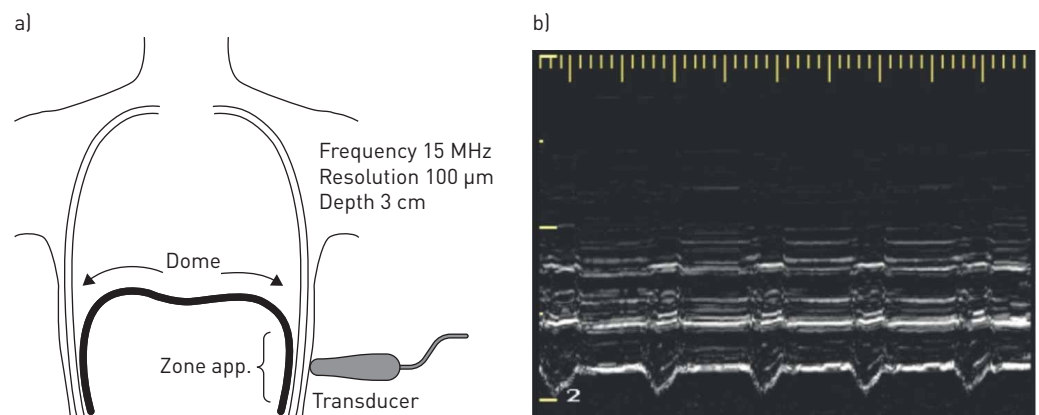


FIGURE 5 Diaphragm ultrasound assessment. a) When measuring diaphragm thickness and thickening fraction, the use of a linear, high-frequency probe is suggested. The probe is positioned in the sagittal-oblique position at the level of the zone of apposition, and image scanning begins at the mid-axillary line. When evaluating diaphragm excursion, use of a curvilinear, low-frequency probe is preferable. The probe is positioned in the sub-hepatic region, with the beam oriented cephalad and posteriorly, aiming at the most cephalad aspect of the diaphragmatic dome. b) M-mode image of diaphragm thickening during inspiration. End-expiratory and end-inspiratory diaphragm thicknesses can be directly measured and thickening fraction can be determined.

thickening explain only one third (or less) of the variability in inspiratory effort [180, 187, 188]. This is not surprising, considering that thickening is a one-dimensional measurement, whereas inspiratory effort results from an active three-dimensional displacement of muscle volume. In addition, the extent of diaphragm thickening for a given level of inspiratory effort varies considerably between subjects and the reproducibility of the measurement is weak (reproducibility coefficients range from 16% to 27%) [187].

In critically ill patients receiving pressure support ventilation, TF <29% has been associated with diaphragm dysfunction, the latter being defined as $P_{ao,tw} < 11 \text{ cmH}_2\text{O}$ [181]. In addition, diaphragm TF moderately correlates to indices of neural respiratory drive such as $P_{0.1}$ [188] and has been reported as a possible predictor of weaning outcome [181, 182, 189] and duration of mechanical ventilation [181, 189]. In patients with acute exacerbation of COPD, preliminary data suggests that TF is related to failure of noninvasive ventilation and mortality [190]. Measurements of expiratory and inspiratory diaphragm thickness can be performed using either B- or M-mode ultrasound. The use of M-mode offers the theoretical advantage of making the recording of both variables in a single inspiratory/expiratory cycle easier, and the manual measurement of diaphragm thickness on the same ultrasound frozen image. Whether this translates into a clinically significant difference in measurement compared with B-mode remains to be determined. No studies have yet evaluated TF or TR during exercise.

3.1.3. Diaphragm excursion

Excursion of the right diaphragm has high intra- and inter-observer reliability [191, 192] and its lower limit of normal is 3.6 cm in women and 4.7 cm in men during maximal inspiratory efforts [191]. From a technical point of view, measurement errors may occur when the displacement of the diaphragm is not optimally aligned with the M-mode plane, but angle-independent M-mode sonography may mitigate this effect [193].

Diaphragm excursion is sensitive to changes in respiratory pattern [194], is related to the volume-generating capacity of the diaphragm (measured using VC) following abdominal surgery [195] and has been used to identify diaphragm weakness in the setting of acute exacerbation of COPD [196] and acute stroke [197]. In intubated patients, diaphragm excursion is moderately related to P_{di} [198] and, possibly, to weaning outcome [192, 199]. In children, ultrasound imaging has been used to assess anatomical defects of the diaphragm (lobulated-shaped hemidiaphragms, focal diaphragmatic eventration, diaphragmatic hernia) and to document paradoxical movements of the diaphragm (supplementary material) [200].

3.2. Optoelectronic plethysmography

Optoelectronic plethysmography (OEP) is an established technique that allows tidal changes in the volume of the chest wall and its compartments to be measured (figure 6) [201, 202]. By using this technique, investigators reported that patients with more severe COPD consistently experience dynamic hyperinflation during incremental exercise, while other patients, specifically those with a greater expiratory flow reserve at rest, adopted at least two significantly different patterns of change in end-expiratory volume of the chest wall [203–205]: some showed a progressive significant increase in end-expiratory volume of the chest wall (“early hyperinflators”) and others showed an increase only at higher levels of exercise (“late hyperinflators”). Three different, distinct patterns of breathing and chest wall volume displacement were found in patients with severe COPD, interstitial pulmonary fibrosis and cystic fibrosis to cope with chronic respiratory failure [206].

OEP has been used to evaluate a variety of NMDs, such as Duchenne [207–209], limb girdle and Becker muscular dystrophies, facioscapulohumeral dystrophy [210] and ALS [211]. OEP has also been used to assess the effects of different surgical techniques (such as laparoscopic surgery) on chest wall kinematics and inspiratory muscle activity [212], Nuss technique for pectus excavatum [213], diaphragm plication for unilateral diaphragm paralysis [214], and diaphragm repair in congenital diaphragmatic hernia [215]. More recently, OEP has been used to evaluate the effects on chest wall kinematics of several interventions, such as air stacking [216], breath stacking [217], stretching [218], incentive spirometry [219], inspiratory loaded breathing [220] and rehabilitation [221]. OEP can be used to monitor tidal breathing and respiratory muscle function in newborns [222], in children with spinal muscle atrophy type 1 and type 2 [223] and in children and young adults with Duchenne muscular dystrophy [207, 208].

3.3. Other investigations

Chest radiography and computed tomography have been used to assess the position of the diaphragm, particularly to identify diaphragm elevation secondary to weakness or paralysis in patients with myopathies, neuropathies and injured hemidiaphragm [224]. Chest fluoroscopy, although highly ionising, has been used to identify decreased or paradoxical diaphragm motion.

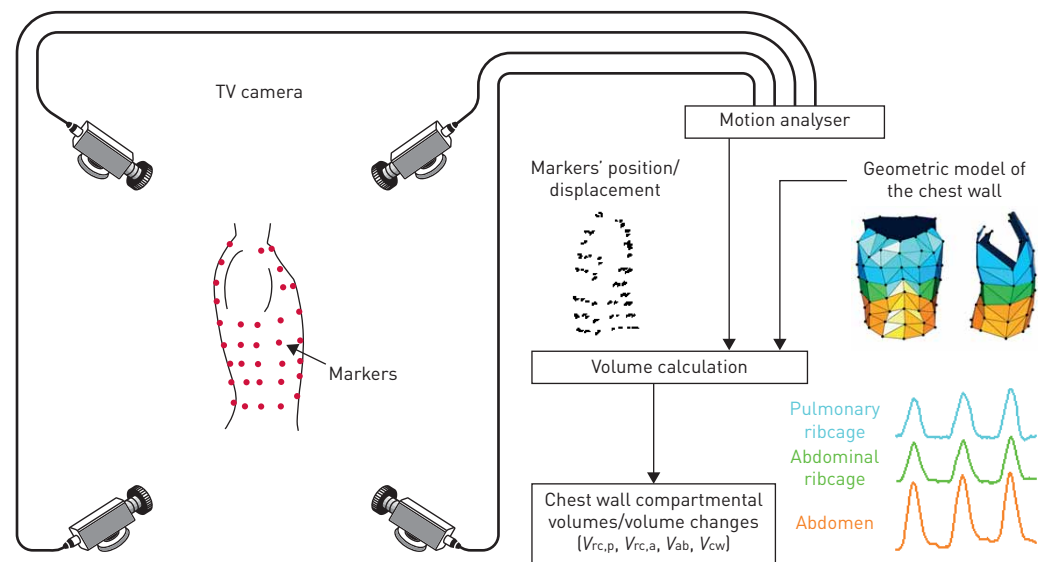


FIGURE 6 Optoelectronic plethysmography. A number of reflective markers are positioned on the trunk of the subject in selected anatomical reference sites of the ribcage and the abdomen. A set of cameras placed nearby the subject under analysis and dedicated stereo-photogrammetric techniques allow measuring the position (three-dimensional coordinates) and motion of the markers. A closed surface is defined by connecting the points and the volume enclosed by the thoraco-abdominal surface and its different parts is computed using Gauss' theorem. The chest wall is typically modelled as being composed of three different compartments: pulmonary ribcage (rc,p), exposed on its inner surface to pleural pressure, abdominal ribcage (rc,a), and the abdomen (ab), the latter both exposed to abdominal pressure. Total chest wall volume (V_{cw}) is the sum of the volume of these three compartments ($V_{rc,p}$, $V_{rc,a}$ and V_{ab}).

Two- and three-dimensional magnetic resonance imaging (MRI) is being increasingly used, particularly in neuromuscular diseases [224], to assess muscle size, structure and altered function by using different tissue-weighting (T1, T2 and proton density). Two-dimensional MRI can assess qualitatively muscular atrophy on axial and coronal images and measure the cranio-caudal diaphragm movement. Dynamic MRI provides information on the motion of the chest wall and the diaphragm on sagittal images [225]. Given the paucity of published studies on this topic, it is difficult to draw conclusions on this imaging tool; further studies are needed to evaluate the validity, precision, reproducibility, prognostic value and responsiveness to interventions of dynamic MRI of the diaphragm.

Structured light plethysmography (SLP) is another emerging imaging tool. SLP is a non-contact, noninvasive method to assess breathing pattern [226]. The technique is based on the stereoscopic analysis of respiratory-related distortions of a black and white chequered pattern projected on the chest wall and abdomen [226–228]. SLP has been validated in healthy subjects and in patients [226–229]. In a recent study, NIERAT *et al.* [226] reported that SLP can detect differences in breathing pattern in COPD compared with healthy controls. In the same study, SLP allowed measurement of ventilatory activity while preserving resting tidal breathing variability, reducing instrumental observer effect and avoiding any disruptions in breathing pattern induced by the use of the pneumotachograph-mouthpiece-noseclip combination. SLP allows a detailed compartmentalised analysis of thoraco-abdominal behaviour, which is not the case for wearable devices [226]. In children with asthma, SLP can differentiate those with and without airway obstruction, and may identify responses to bronchodilator [230]. Further research is, however, required to confirm the clinical applications of SLP.

Section 4. Respiratory muscle structure, perfusion and metabolism

Several methodological approaches can provide a comprehensive assessment of the mechanisms regulating respiratory muscle blood flow and oxygen delivery in relation to oxidative metabolic demand and mitochondrial function, as well as the consequences of oxidative stress and inflammation (table 6). These techniques have the potential to be used for monitoring interventions aimed at restoring respiratory muscle function in the ICU and the rehabilitation setting.

4.1. Near-infrared spectroscopy

A decade ago, a technique combining near-infrared spectroscopy (NIRS) with the light absorbing tracer dye indocyanine green (ICG) was employed to measure intercostal muscle blood flow (IMBF) using Fick's

TABLE 6 Laboratory techniques for evaluation of respiratory muscle structure, perfusion and metabolism

Techniques	Invasiveness	Physiology laboratory required	Biology laboratory required	Purpose
Near-infrared spectroscopy	None	Yes	No	Muscle blood flow
Oxygen cost of breathing	None	Yes	No	Ventilation, oxygen uptake
Access to costal diaphragm muscle	Yes, thoracotomy	Yes, always in surgery room	No	Biological and histological analyses
Access to parasternal muscles	Yes, thoracotomy	Yes, always in surgery room	No	Biological and histological analyses
Access to external intercostals	Yes, open biopsy techniques	Yes, possible in surgery room	No	Biological and histological analyses
Immunohistochemical or immunofluorescence analyses		No	Yes	Muscle fibre type and morphometry
Mitochondrial respiratory chain evaluation (respiration procedures)	None	No	Yes	Quantification of mitochondrial respiration (oxygen consumption)
Immunoblotting procedures	None	No	Yes	Quantification of protein levels in muscle specimens
Quantitative real-time PCR	None	No	Yes	Quantification of gene expression levels in muscle specimens
Specific activity assays including mitochondrial enzyme activities	None	No	Yes	Quantification of activity levels of enzymes in muscle specimens

principle. GUENETTE *et al.* [231] were the first to quantify IMBF in healthy subjects during resting isocapnic hyperpnoea at different fractions of MVV. They reported that as ventilation rose, IMBF significantly correlated with the increase in cardiac output, WOB and P_{di} , suggesting that the NIRS-ICG technique is a sensitive indicator of IMBF in healthy humans. Similar results have been reported by Vogiatzis and co-workers, employing the same NIRS-ICG technique to measure IMBF in healthy subjects [232] and COPD patients [233].

Absolute IMBF measurements *via* the NIRS-ICG technique require arterial cannulation. For this reason, an alternative method was proposed to measure relative changes in muscle perfusion from rest, namely the blood flow index (BFI), requiring only venous catheterisation for the injection of ICG (figure 7) [234].

HABAZETTL *et al.* [234] compared BFI values obtained from the seventh intercostal space against absolute muscle blood flow determined using the NIRS-ICG technique during cycling in healthy subjects. The investigators reported a very good agreement between BFI and NIRS-ICG techniques in healthy subjects during cycling [234], and also (by retrospective data analysis) in patients with COPD (figure 7) [233]. GUENETTE *et al.* [235] showed that BFI of intercostal and sternocleidomastoid muscles during isocapnic hyperpnoea was strongly correlated with WOB and surface EMG, thus confirming that the BFI technique provides a minimally invasive and technically less demanding alternative to NIRS-ICG for measuring respiratory muscle perfusion in humans at rest and during exercise.

4.2. Oxygen cost of breathing

The oxygen cost of breathing is an index of the energy required for ventilation. For more detailed information on methods of assessment please refer to the supplementary material. Oxygen cost of breathing was shown to be increased in women [236, 237] and in obesity [238, 239], post-operative patients [240], COPD [240, 241], cystic fibrosis [242, 243], children with asthma [244], sarcoidosis [245] and chronic heart failure [246, 247]. In these conditions, the increased oxygen cost of breathing may contribute to increased energy cost during activities of daily living adding, particularly in diseases imposing a ventilator or cardiac constraint, an extra contribution to the reduced exercise capacity. Several interventions have been used in different patient populations to reduce WOB, which has a potential impact on reducing the oxygen cost of breathing, namely, invasive and noninvasive ventilation [248–250], high flow nasal oxygen [248, 251], ventilation with heliox [252], respiratory muscle training [253, 254] and exercise training [255].

4.3. Biopsy (specificities for respiratory muscles)

In recent years, respiratory muscles have been studied through the analyses of the costal diaphragm, with very restricted access, and only *via* thoracotomy performed for clinical reasons (mainly lung cancer and lung volume reduction surgeries). During thoracotomy, parasternal and diaphragm biopsy specimens have been obtained from the third interspace and the anterior costal diaphragm lateral to the insertion of the phrenic nerve, respectively [256–260]. Additionally, other studies have been based on the analysis of the

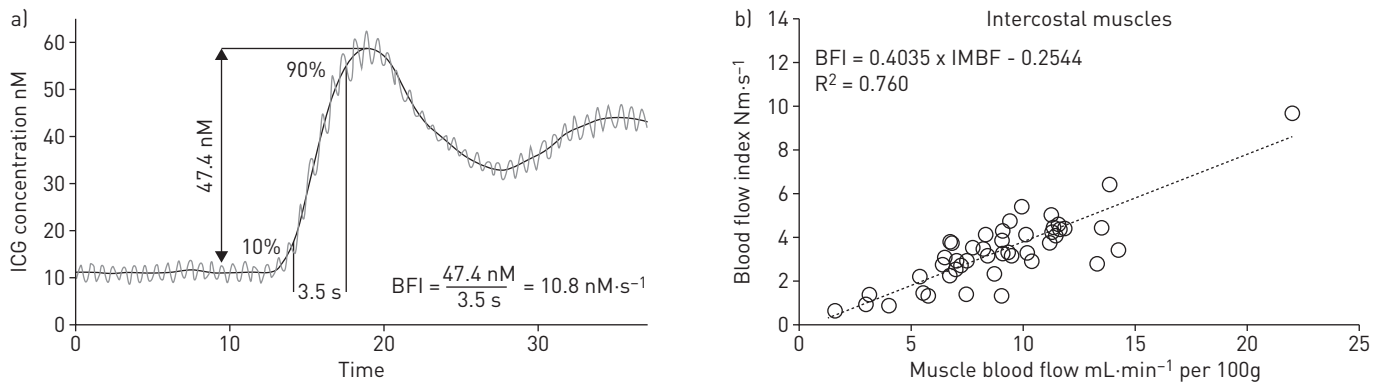


FIGURE 7 Near-infrared spectroscopy (NIRS). a) Typical example of muscle indocyanine green (ICG) concentration curve recorded by NIRS during exercise. The original tracing (grey line) appears with marked oscillations (at a frequency of 84 min^{-1} ; 1.4 Hz) due to muscle contraction and relaxation during cycling. Low-pass filtering with a cut-off frequency of 0.5 Hz produced the smoothed curve (black line) that was used for blood flow index (BFI) calculation. Data points at 10 and 90% of ICG concentration peak are indicated, and an example of BFI calculation is given. Reproduced with permission from the publisher [234]. b) Regression analysis of individual BFI assessed by the NIRS-ICG method *versus* actually measured muscle blood flow assessed by Fick's principle at different levels of minute ventilation recorded during isocapnic hyperpnoea trials for the intercostal muscles in chronic obstructive pulmonary disease. IMBF: intercostal muscle blood flow. Data calculated from [233].

external intercostal muscle following procedures involving an open biopsy technique [257, 258, 261–265]. Biopsies from the external intercostals have usually been taken along the anterior axillary line at the sixth intercostal space, as detailed in previous studies [257, 258, 261–265]. In patients with chronic respiratory conditions, limb muscles are more severely affected than respiratory muscles, which need to overcome the increased inspiratory loads and may exhibit adaptive features [266].

4.4. Typology

Respiratory muscles undergo a series of structural changes in lung diseases. These changes have been extensively studied in patients with COPD, in which the diaphragm shows increased type I fibres [267], favouring aerobic metabolism [268]. Structural changes in the respiratory muscles (injury and regeneration cycles) depend mainly on the effect derived from increased ventilatory loads [269, 270]. Increases in capillary and mitochondria numbers and sarcomere length have also been demonstrated [271], along with sarcomere and sarcomeric damage and greater friability of diaphragm [272]. Diaphragm atrophy in patients with COPD has been reported by some, but not all, investigators [256, 259, 273, 274]. Changes in the proportions of fibre types have also been observed in the parasternal and external intercostal muscles of COPD patients [257, 275]. In the latter muscles, an increase in capillary numbers was also described [276], together with fibre atrophy [277]. Respiratory muscle training increased fibre sizes and proportions of type I fibres [278]. In OSAS patients, increased proportions of type I fibres have been reported in the intercostal muscles [262], while no data is available for the diaphragm. Prolonged mechanical ventilation induces sarcomere damage and fibre atrophy in the diaphragm, with no relevant changes in fibre type proportions [279, 280].

4.5. Mitochondrial function

In rats, mitochondrial respiratory rates are lower in the diaphragm than in peripheral muscles [281]. In mice, hypoxia differentially affected peripheral and respiratory muscles with decreased mitochondrial content due to reduced mitochondrial biogenesis and increased mitophagy [282].

Mitochondrial function is altered in patients with COPD [260, 283, 284]. In these patients, mitochondria isolated from intercostal muscles demonstrate electron transport blockade and excessive production of reactive oxygen species, similar to effects found in the vastus lateralis [259, 284]. In the diaphragm, overall mitochondrial respiratory chain capacity was increased and had a higher efficiency in patients with moderate [285] and severe [286] COPD than in healthy controls. In patients with COPD the oxidative capacity of the diaphragm is greater than that of the peripheral muscles [258].

Mitochondrial function and content are impaired in patients with sepsis [287]. In turn, animal models of prolonged mechanical ventilation demonstrate only minor changes in oxidative phosphorylation coupling in diaphragmatic mitochondria [288]. Attempts to improve mitochondrial function using anabolic steroids failed in a hamster model of emphysema [289]. Increased mitochondrial enzyme activity was shown in rodent diaphragm in response to endurance training [290].

4.6. Oxidative stress

Increased oxidant production has been reported in mitochondria and membrane compartments of diaphragm fibres in patients with severe COPD [256, 259]. In several studies [256, 259, 291, 292], the diaphragm of these patients exhibited increased levels of oxidative stress. Such levels inversely correlated with global respiratory and diaphragm muscle function among patients with more severe disease [256, 259]. Contractile actin and myosin, creatine kinase, and carbonic anhydrase-3 are oxidatively modified in the diaphragm of patients with severe COPD, while protein content of myosin [259, 291, 292] and creatine kinase and its activity are reduced [259]. Nonetheless, in saponin-skinned diaphragm and intercostal muscle fibres [265, 286], creatine kinase activity levels do not differ between severe COPD and healthy controls. In external intercostals of COPD patients [264] and in those with OSAS, oxidative stress levels are increased and treatment with continuous positive airway pressure for 6 months does not reduce those levels [262]. In external intercostal muscles of patients with severe sepsis, oxidative stress levels do not differ from those in controls [264]. Oxidative stress in the diaphragm of critically ill patients receiving mechanical ventilation is increased compared to controls [280, 293, 294]. In elderly subjects, markers of oxidative stress are increased in the external intercostals compared to young controls [261].

4.7. Inflammation

Systemic inflammation is a contributor of muscle dysfunction in COPD [295]. In contrast, local inflammation does not play a role in COPD muscle dysfunction: inflammatory cell counts were very low in the diaphragm and external intercostals of patients with severe COPD with preserved body composition [257]. Expression of mRNA and protein content of tumour necrosis factor alpha and interleukin-6 are greater in the external intercostals of patients with severe COPD and normal weight than in healthy controls, while muscle mRNA levels of CD18 panleukocyte marker do not differ between patients and controls [296]. In patients with severe sepsis, inflammatory markers are increased in the external intercostals compared to controls [263].

Collectively, respiratory muscle dysfunction in patients with COPD is the result of multiple deleterious factors such as lung hyperinflation (mechanical disadvantage), gas exchange abnormalities, impaired bioenergetics (increased cost of breathing) and biological mechanisms (oxidative stress), and structural abnormalities (sarcomere damage and atrophy), while inflammation does not seem to play a major role [295]. This scenario coexists with adaptive features including a switch towards a more oxidative phenotype (predominance of slow-twitch fibres, increased mitochondrial density and myoglobin content), probably in response to increased mechanical loads.

Conclusion

Respiratory muscle dysfunction is a major clinical concern in a variety of conditions, from respiratory diseases to NMDs, critically ill patients, sports medicine and paediatric populations. Assessment of respiratory muscle function is therefore of critical importance for patient diagnosis, follow-up and for evaluating the effect of therapeutic interventions aimed at improving respiratory function. 17 years after the 2002 ATS/ERS statement on respiratory muscle testing [1], a growing body of literature has emerged and has been discussed in this document, which provides clinicians and investigators with the latest knowledge on this topic. In addition to historical evidence on respiratory muscle strength, endurance and fatigue assessments, new information on imaging technologies and respiratory muscle assessment during exercise have provided important insights into respiratory muscle function, including its integration with brain and cardiovascular function, dyspnoea and exercise tolerance. This document, which has involved experts in the field of respiratory medicine and physiology on the topic of respiratory muscle testing at rest and during exercise, is intended to open up new perspectives in both clinical and research settings. Despite the remarkable advances in respiratory muscle and lung mechanics assessment in the past few decades, this body of knowledge has not been fully translated to the clinical care of individual patients. Although this state of affairs is likely explained by multiple reasons, it is noteworthy that less and less time has been devoted to training in the administration and interpretation of the more advanced tests of respiratory muscle function worldwide. This contributes to a vicious circle in which fewer pulmonologists master the use of less common devices that are available only in specialised centres. To fight this regrettable situation, it seems apparent that new generations of pulmonologists should (again) be intensively exposed to clinical physiology concepts and practices. To reach this intent, the key relevance of the leading societies in our field (*e.g.* the European Respiratory Society, American Thoracic Society and American College of Chest Physicians) cannot be underestimated.

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References

- 1 American Thoracic Society/European Respiratory Society. ATS/ERS statement on respiratory muscle testing. *Am J Respir Crit Care Med* 2002; 166: 518–624.
- 2 Loring SH, Yoshino K, Kimball WR, *et al.* Gravitational and shear-associated pressure gradients in the abdomen. *J Appl Physiol* 1994; 77: 1375–1382.
- 3 Jackson AC, Vinegar A. A technique for measuring frequency response of pressure, volume, and flow transducers. *J Appl Physiol Respir Environ Exerc Physiol* 1979; 47: 462–467.
- 4 Milic-Emili J, Mead J, Turner JM, *et al.* Improved technique for estimating pleural pressure from esophageal balloons. *J Appl Physiol* 1964; 19: 207–211.
- 5 Walterspercher S, Isaak L, Guttmann J, *et al.* Assessing respiratory function depends on mechanical characteristics of balloon catheters. *Respir Care* 2014; 59: 1345–1352.
- 6 Augusto RM, Albuquerque AL, Jaeger T, *et al.* Stability and agreement of a microtransducer and an air-filled balloon esophageal catheter in the monitoring of esophageal pressure. *Respir Care* 2017; 62: 215–221.
- 7 Heritier F, Rahm F, Pasche P, *et al.* Sniff nasal inspiratory pressure. A noninvasive assessment of inspiratory muscle strength. *Am J Respir Crit Care Med* 1994; 150: 1678–1683.
- 8 Mead J. Mechanical properties of lungs. *Physiol Rev* 1961; 41: 281–330.
- 9 Killian KJ, Jones NL. Respiratory muscles and dyspnea. *Clin Chest Med* 1988; 9: 237–248.
- 10 Rodrigues A, Da Silva ML, Berton DC, *et al.* Maximal inspiratory pressure: does the choice of reference values actually matter? *Chest* 2017; 152: 32–39.
- 11 Sclausser Pessoa IM, Franco Parreira V, Fregonezi GA, *et al.* Reference values for maximal inspiratory pressure: a systematic review. *Can Respir J* 2014; 21: 43–50.
- 12 Maillard JO, Burdet L, van Melle G, *et al.* Reproducibility of twitch mouth pressure, sniff nasal inspiratory pressure, and maximal inspiratory pressure. *Eur Respir J* 1998; 11: 901–905.
- 13 Nikolettou D, Rafferty G, Man WD, *et al.* Sniff nasal inspiratory pressure in patients with moderate-to-severe chronic obstructive pulmonary disease: learning effect and short-term between-session repeatability. *Respiration* 2014; 88: 365–370.
- 14 Khirani S, Colella M, Caldarelli V, *et al.* Longitudinal course of lung function and respiratory muscle strength in spinal muscular atrophy type 2 and 3. *Eur J Paediatr Neurol* 2013; 17: 552–560.
- 15 King M, Brock G, Lundell C. Clearance of mucus by simulated cough. *J Appl Physiol* 1985; 58: 1776–1782.
- 16 Suarez AA, Pessolano FA, Monteiro SG, *et al.* Peak flow and peak cough flow in the evaluation of expiratory muscle weakness and bulbar impairment in patients with neuromuscular disease. *Am J Phys Med Rehab* 2002; 81: 506–511.
- 17 Sancho J, Servera E, Diaz J, *et al.* Comparison of peak cough flows measured by pneumotachograph and a portable peak flow meter. *Am J Phys Med Rehab* 2004; 83: 608–612.
- 18 Bach JR. Update and perspectives on noninvasive respiratory muscle aids 0.1. The inspiratory aids. *Chest* 1994; 105: 1230–1240.
- 19 Tzani P, Chiesa S, Aiello M, *et al.* The value of cough peak flow in the assessment of cough efficacy in neuromuscular patients. A cross sectional study. *Eur J Phys Rehab Med* 2014; 50: 427–432.
- 20 Steier J, Kaul S, Seymour J, *et al.* The value of multiple tests of respiratory muscle strength. *Thorax* 2007; 62: 975–980.
- 21 Man WDC, Kyroussis D, Fleming TA, *et al.* Cough gastric pressure and maximum expiratory mouth pressure in humans. *Am J Respir Crit Care Med* 2003; 168: 714–717.
- 22 Koulouris N, Mulvey DA, Laroche CM, *et al.* The measurement of inspiratory muscle strength by sniff esophageal, nasopharyngeal, and mouth pressures. *Am Rev Respir Dis* 1989; 139: 641–646.
- 23 Garcia-Rio F, Mediano O, Pino JM, *et al.* Noninvasive measurement of the maximum relaxation rate of inspiratory muscles in patients with neuromuscular disorders. *Respiration* 2006; 73: 474–480.
- 24 Luo YM, Hart N, Mustfa N, *et al.* Reproducibility of twitch and sniff transdiaphragmatic pressures. *Respir Physiol Neurobiol* 2002; 132: 301–306.
- 25 Mulvey DA, Elliott MW, Koulouris NG, *et al.* Sniff esophageal and nasopharyngeal pressures and maximal relaxation rates in patients with respiratory dysfunction. *Am Rev Respir Dis* 1991; 143: 950–953.
- 26 Evans SA, Watson L, Hawkins M, *et al.* Respiratory muscle strength in chronic heart failure. *Thorax* 1995; 50: 625–628.
- 27 Ward K, Seymour J, Steier J, *et al.* Acute ischaemic hemispheric stroke is associated with impairment of reflex in addition to voluntary cough. *Eur Respir J* 2010; 36: 1383–1390.

- 28 Fauroux B, Aubertin G, Cohen E, *et al.* Sniff nasal inspiratory pressure in children with muscular, chest wall or lung disease. *Eur Respir J* 2009; 33: 113–117.
- 29 Aiello M, Rampello A, Granella F, *et al.* Cough efficacy is related to the disability status in patients with multiple sclerosis. *Respiration* 2008; 76: 311–316.
- 30 Nicot F, Hart N, Forin V, *et al.* Respiratory muscle testing: a valuable tool for children with neuromuscular disorders. *Am J Respir Crit Care Med* 2006; 174: 67–74.
- 31 Quijano-Roy S, Khirani S, Colella M, *et al.* Diaphragmatic dysfunction in collagen VI myopathies. *Neuromuscul Disord* 2014; 24: 125–133.
- 32 Morgan RK, McNally S, Alexander M, *et al.* Use of sniff nasal-inspiratory force to predict survival in amyotrophic lateral sclerosis. *Am J Respir Crit Care Med* 2005; 171: 269–274.
- 33 Polkey MI, Lyall RA, Yang K, *et al.* Respiratory muscle strength as a predictive biomarker for survival in amyotrophic lateral sclerosis. *Am J Respir Crit Care Med* 2017; 195: 86–95.
- 34 Criner G, Cordova FC, Leyenson V, *et al.* Effect of lung volume reduction surgery on diaphragm strength. *Am J Respir Crit Care Med* 1998; 157: 1578–1585.
- 35 Kyrrousis D, Polkey MI, Keilty SE, *et al.* Exhaustive exercise slows inspiratory muscle relaxation rate in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 1996; 153: 787–793.
- 36 De Troyer A, Borenstein S, Cordier R. Analysis of lung volume restriction in patients with respiratory muscle weakness. *Thorax* 1980; 35: 603–610.
- 37 Fallat RJ, Jewitt B, Bass M, *et al.* Spirometry in amyotrophic lateral sclerosis. *Arch Neurol* 1979; 36: 74–80.
- 38 Lechtzin N, Wiener CM, Shade DM, *et al.* Spirometry in the supine position improves the detection of diaphragmatic weakness in patients with amyotrophic lateral sclerosis. *Chest* 2002; 121: 436–442.
- 39 Varrato J, Siderowf A, Damiano P, *et al.* Postural change of forced vital capacity predicts some respiratory symptoms in ALS. *Neurology* 2001; 57: 357–359.
- 40 Lisboa C, Pare PD, Pertuze J, *et al.* Inspiratory muscle function in unilateral diaphragmatic paralysis. *Am Rev Respir Dis* 1986; 134: 488–492.
- 41 Laroche CM, Mier AK, Moxham J, *et al.* Diaphragm strength in patients with recent hemidiaphragm paralysis. *Thorax* 1988; 43: 170–174.
- 42 Laroche CM, Carroll N, Moxham J, *et al.* Clinical significance of severe isolated diaphragm weakness. *Am Rev Respir Dis* 1988; 138: 862–866.
- 43 Mier-Jedrzejowicz A, Brophy C, Moxham J, *et al.* Assessment of diaphragm weakness. *Am Rev Respir Dis* 1988; 137: 877–883.
- 44 Stambler N, Charatan M, Cedarbaum JM. Prognostic indicators of survival in ALS. ALS CNTF Treatment Study Group. *Neurology* 1998; 50: 66–72.
- 45 Lacomblez L, Bensimon G, Leigh PN, *et al.* Dose-ranging study of riluzole in amyotrophic lateral sclerosis. Amyotrophic Lateral Sclerosis/Riluzole Study Group II. *Lancet* 1996; 347: 1425–1431.
- 46 Meininger V, Bensimon G, Bradley WR, *et al.* Efficacy and safety of xaliproden in amyotrophic lateral sclerosis: results of two phase III trials. *Amyotroph Lateral Scler Other Motor Neuron Disord* 2004; 5: 107–117.
- 47 Phillips MF, Quinlivan RC, Edwards RH, *et al.* Changes in spirometry over time as a prognostic marker in patients with Duchenne muscular dystrophy. *Am J Respir Crit Care Med* 2001; 164: 2191–2194.
- 48 Chevrolet JC, Deleamont P. Repeated vital capacity measurements as predictive parameters for mechanical ventilation need and weaning success in the Guillain-Barre syndrome. *Am Rev Respir Dis* 1991; 144: 814–818.
- 49 Bourke SC, Tomlinson M, Williams TL, *et al.* Effects of non-invasive ventilation on survival and quality of life in patients with amyotrophic lateral sclerosis: a randomised controlled trial. *Lancet Neurol* 2006; 5: 140–147.
- 50 Van der Beek NA, Hagemans ML, Reuser AJ, *et al.* Rate of disease progression during long-term follow-up of patients with late-onset Pompe disease. *Neuromuscul Disord* 2009; 19: 113–117.
- 51 Clinical indications for noninvasive positive pressure ventilation in chronic respiratory failure due to restrictive lung disease, COPD, and nocturnal hypoventilation – a consensus conference report. *Chest* 1999; 116: 521–534.
- 52 Johnson EM, Roberts M, Mozaffar T, *et al.* Pulmonary function tests (maximum inspiratory pressure, maximum expiratory pressure, vital capacity, forced vital capacity) predict ventilator use in late-onset Pompe disease. *Neuromuscul Disord* 2016; 26: 136–145.
- 53 Ragette R, Mellies U, Schwake C, *et al.* Patterns and predictors of sleep disordered breathing in primary myopathies. *Thorax* 2002; 57: 724–728.
- 54 Akoumianaki E, Maggiore SM, Valenza F, *et al.* The application of esophageal pressure measurement in patients with respiratory failure. *Am J Respir Crit Care Med* 2014; 189: 520–531.
- 55 Kyrrousis D, Mills GH, Polkey MI, *et al.* Abdominal muscle fatigue after maximal ventilation in humans. *J Appl Physiol* 1996; 81: 1477–1483.
- 56 Hamnegard CH, Wrang S, Kyrrousis D, *et al.* Diaphragm fatigue following maximal ventilation in man. *Eur Respir J* 1996; 9: 241–247.
- 57 Maltais F, Reissmann H, Gottfried SB. Pressure support reduces inspiratory effort and dyspnea during exercise in chronic airflow obstruction. *Am J Respir Crit Care Med* 1995; 151: 1027–1033.
- 58 Sassoon CS, Lodia R, Light RW, *et al.* Maximum inspiratory muscle endurance capacity during resistive loading in chronic obstructive pulmonary disease. *Respiration* 1990; 57: 343–350.
- 59 Polkey MI, Kyrrousis D, Hamnegard CH, *et al.* Diaphragm performance during maximal voluntary ventilation in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 1997; 155: 642–648.
- 60 Field S, Sancu S, Grassino A. Respiratory muscle oxygen consumption estimated by the diaphragm pressure-time index. *J Appl Physiol Respir Environ Exerc Physiol* 1984; 57: 44–51.
- 61 Jubran A, Tobin MJ. Pathophysiologic basis of acute respiratory distress in patients who fail a trial of weaning from mechanical ventilation. *Am J Respir Crit Care Med* 1997; 155: 906–915.
- 62 Laveneziana P, Webb KA, Wadell K, *et al.* Does expiratory muscle activity influence dynamic hyperinflation and exertional dyspnea in COPD? *Respir Physiol Neurobiol* 2014; 199: 24–33.
- 63 Duranti R, Bonetti L, Vivoli P, *et al.* Dyspnea during exercise in hyperbaric conditions. *Med Sci Sports Exerc* 2006; 38: 1932–1938.
- 64 Faisal A, Alghamdi BJ, Ciavaglia CE, *et al.* Common mechanisms of dyspnea in chronic interstitial and obstructive lung disorders. *Am J Respir Crit Care Med* 2016; 193: 299–309.

- 65 O'Donnell DE, Laveneziana P, Webb K, *et al.* Chronic obstructive pulmonary disease: clinical integrative physiology. *Clin Chest Med* 2014; 35: 51–69.
- 66 Laveneziana P, Webb KA, Ora J, *et al.* Evolution of dyspnea during exercise in chronic obstructive pulmonary disease: impact of critical volume constraints. *Am J Respir Crit Care Med* 2011; 184: 1367–1373.
- 67 Kushida CA. The use of esophageal manometry in the diagnosis of sleep-related breathing disorders. *Conf Proc IEEE Eng Med Biol Soc* 2004; 5: 3860–3863.
- 68 Jubran A, Grant BJ, Laghi F, *et al.* Weaning prediction: esophageal pressure monitoring complements readiness testing. *Am J Respir Crit Care Med* 2005; 171: 1252–1259.
- 69 Petrof BJ, Calderini E, Gottfried SB. Effect of CPAP on respiratory effort and dyspnea during exercise in severe COPD. *J Appl Physiol* 1990; 69: 179–188.
- 70 Eves ND, Petersen SR, Haykowsky MJ, *et al.* Helium-hyperoxia, exercise, and respiratory mechanics in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2006; 174: 763–771.
- 71 Hatipoglu U, Laghi F, Tobin M. Does inhaled albuterol improve diaphragmatic contractility in patients with chronic obstructive pulmonary disease? *Am J Respir Crit Care Med* 1999; 160: 1916–1921.
- 72 Man WD, Mustafa N, Nikolettou D, *et al.* Effect of salmeterol on respiratory muscle activity during exercise in poorly reversible COPD. *Thorax* 2004; 59: 471–476.
- 73 O'Donnell DE, Hamilton AL, Webb K. Sensory-mechanical relationships during high-intensity, constant-work-rate exercise in COPD. *J Appl Physiol* 2006; 101: 1025–1035.
- 74 Collins EG, Langbein WE, Fehr L, *et al.* Can ventilation-feedback training augment exercise tolerance in patients with chronic obstructive pulmonary disease? *Am J Respir Crit Care Med* 2008; 177: 844–852.
- 75 Macklem PT. Therapeutic implications of the pathophysiology of COPD. *Eur Respir J* 2010; 35: 676–680.
- 76 Spahija J, de Marchie M, Grassino A. Effects of imposed pursed-lips breathing on respiratory mechanics and dyspnea at rest and during exercise in COPD. *Chest* 2005; 128: 640–650.
- 77 Breslin EH. The pattern of respiratory muscle recruitment during pursed-lip breathing. *Chest* 1992; 101: 75–78.
- 78 Hughes PD, Polkey MI, Kyroussis D, *et al.* Measurement of sniff nasal and diaphragm twitch mouth pressure in patients. *Thorax* 1998; 53: 96–100.
- 79 Luo YM, Hart N, Mustafa N, *et al.* Reproducibility of twitch and sniff transdiaphragmatic pressures. *Respir Physiol Neurobiol* 2002; 132: 301–306.
- 80 Laghi F, Cattapan SE, Jubran A, *et al.* Is weaning failure caused by low-frequency fatigue of the diaphragm? *Am J Respir Crit Care Med* 2003; 167: 120–127.
- 81 Rafferty GF, Greenough A, Dimitriou G, *et al.* Assessment of neonatal diaphragm function using magnetic stimulation of the phrenic nerves. *Am J Respir Crit Care Med* 2000; 162: 2337–2340.
- 82 Dimitriou G, Greenough A, Moxham J, *et al.* Influence of maturation on infant diaphragm function assessed by magnetic stimulation of phrenic nerves. *Pediatr Pulmonol* 2003; 35: 17–22.
- 83 Rafferty GF, Mustafa N, Man WD, *et al.* Twitch airway pressure elicited by magnetic phrenic nerve stimulation in anesthetized healthy children. *Pediatr Pulmonol* 2005; 40: 141–147.
- 84 Supinski GS, Callahan LA. Diaphragm weakness in mechanically ventilated critically ill patients. *Crit Care* 2013; 17: R120.
- 85 Mills GH, Ponte J, Hamnegard CH, *et al.* Tracheal tube pressure change during magnetic stimulation of the phrenic nerves as an indicator of diaphragm strength on the intensive care unit. *Br J Anaesth* 2001; 87: 876–884.
- 86 Langer D, Jacome C, Charususin N, *et al.* Measurement validity of an electronic inspiratory loading device during a loaded breathing task in patients with COPD. *Respir Med* 2013; 107: 633–635.
- 87 Wuthrich TU, Marty J, Benaglia P, *et al.* Acute effects of a respiratory sprint-interval session on muscle contractility. *Med Sci Sports Exerc* 2015; 47: 1979–1987.
- 88 Hart N, Hawkins P, Hamnegard CH, *et al.* A novel clinical test of respiratory muscle endurance. *Eur Respir J* 2002; 19: 232–239.
- 89 Hill K, Jenkins SC, Philippe DL, *et al.* Comparison of incremental and constant load tests of inspiratory muscle endurance in COPD. *Eur Respir J* 2007; 30: 479–486.
- 90 Vincent M, Court-Fortune I, Brun C, *et al.* Determination of normal values for an isocapnic hyperpnea endurance test in healthy individuals. *Respir Physiol Neurobiol* 2016; 230: 5–10.
- 91 Mancini DM, Henson D, La Manca J, *et al.* Benefit of selective respiratory muscle training on exercise capacity in patients with chronic congestive heart failure. *Circulation* 1995; 91: 320–329.
- 92 Mancini DM, Henson D, LaManca J, *et al.* Evidence of reduced respiratory muscle endurance in patients with heart failure. *J Am Coll Cardiol* 1994; 24: 972–981.
- 93 Butler JE. Drive to the human respiratory muscles. *Respir Physiol Neurobiol* 2007; 159: 115–126.
- 94 Luo YM, Moxham J, Polkey MI. Diaphragm electromyography using an oesophageal catheter: current concepts. *Clin Sci* 2008; 115: 233–244.
- 95 Laghi F, Staikh HS, Morales D, *et al.* Diaphragmatic neuromechanical coupling and mechanisms of hypercapnia during inspiratory loading. *Respir Physiol Neurobiol* 2014; 1: 32–41.
- 96 Qin YY, Steier J, Jolley C, *et al.* Efficiency of neural drive during exercise in patients with COPD and healthy subjects. *Chest* 2010; 138: 1309–1315.
- 97 Jolley CJ, Luo YM, Steier J, *et al.* Neural respiratory drive and breathlessness in COPD. *Eur Respir J* 2015; 45: 355–364.
- 98 Schweitzer TW, Fitzgerald JW, Bowden JA, *et al.* Spectral analysis of human inspiratory diaphragmatic electromyograms. *J Appl Physiol Respir Environ Exerc Physiol* 1979; 46: 152–165.
- 99 Bartolo A, Roberts C, Dzwonczyk RR, *et al.* Analysis of diaphragm EMG signals: comparison of gating vs. subtraction for removal of ECG contamination. *J Appl Physiol* 1996; 80: 1898–1902.
- 100 Schmidt M, Chiti L, Hug F, *et al.* Surface electromyogram of inspiratory muscles: a possible routine monitoring tool in the intensive care unit. *Br J Anaesth* 2011; 106: 913–914.
- 101 Gandeia SC, McKenzie DK. Human diaphragmatic EMG: changes with lung volume and posture during supramaximal phrenic stimulation. *J Appl Physiol* 1986; 60: 1420–1428.
- 102 Luo YM, Llyall RA, Lou Harris M, *et al.* Quantification of the esophageal diaphragm electromyogram with magnetic phrenic nerve stimulation. *Am J Respir Crit Care Med* 1999; 160: 1629–1634.

- 103 Jolley CJ, Luo YM, Steier J, *et al.* Neural respiratory drive in healthy subjects and in COPD. *Eur Respir J* 2009; 33: 289–297.
- 104 Reilly CC, Jolley CJ, Ward K, *et al.* Neural respiratory drive measured during inspiratory threshold loading and acute hypercapnia in healthy individuals. *Exp Physiol* 2013; 98: 1190–1198.
- 105 Murphy PB, Kumar A, Reilly C, *et al.* Neural respiratory drive as a physiological biomarker to monitor change during acute exacerbations of COPD. *Thorax* 2011; 66: 602–608.
- 106 MacBean V, Hughes C, Nicol G, *et al.* Measurement of neural respiratory drive *via* parasternal intercostal electromyography in healthy adult subjects. *Physiol Meas* 2016; 37: 2050–2063.
- 107 Suh ES, Mandal S, Harding R, *et al.* Neural respiratory drive predicts clinical deterioration and safe discharge in exacerbations of COPD. *Thorax* 2015; 70: 1123–1130.
- 108 Ramsook AH, Mitchell RA, Bell T, *et al.* Is parasternal intercostal EMG an accurate surrogate of respiratory neural drive and biomarker of dyspnea during cycle exercise testing? *Respir Physiol Neurobiol* 2017; 242: 40–44.
- 109 Similowski T, Mehiri S, Duguet A, *et al.* Comparison of magnetic and electrical phrenic nerve stimulation in assessment of phrenic nerve conduction time. *J Appl Physiol* 1997; 82: 1190–1199.
- 110 Luo YM, Polkey MI, Lyall RA, *et al.* Effect of brachial plexus co-activation on phrenic nerve conduction time. *Thorax* 1999; 54: 765–770.
- 111 Berry RB, Ryals S, Girdhar A, *et al.* Use of chest wall electromyography to detect respiratory effort during polysomnography. *J Clin Sleep Med* 2016; 12: 1239–1244.
- 112 Luo YM, Tang J, Jolley C, *et al.* Distinguishing obstructive from central sleep apnea events: diaphragm electromyogram and esophageal pressure compared. *Chest* 2009; 135: 1133–1141.
- 113 Stoohs RA, Blum HC, Knaack L, *et al.* Comparison of pleural pressure and transcutaneous diaphragmatic electromyogram in obstructive sleep apnea syndrome. *Sleep* 2005; 28: 321–329.
- 114 Luo YM, He BT, Wu YX, *et al.* Neural respiratory drive and ventilation in patients with chronic obstructive pulmonary disease during sleep. *Am J Respir Crit Care Med* 2014; 190: 227–229.
- 115 Elbehairy AF, Guenette JA, Faisal A, *et al.* Mechanisms of exertional dyspnoea in symptomatic smokers without COPD. *Eur Respir J* 2016; 48: 694–705.
- 116 Ciavaglia CE, Guenette JA, Langer D, *et al.* Differences in respiratory muscle activity during cycling and walking do not influence dyspnea perception in obese patients with COPD. *J Appl Physiol* 2014; 117: 1292–1301.
- 117 He BT, Lu G, Xiao SC, *et al.* Coexistence of OSA may compensate for sleep related reduction in neural respiratory drive in patients with COPD. *Thorax* 2017; 72: 256–262.
- 118 De Troyer A, Leeper JB, McKenzie DK, *et al.* Neural drive to the diaphragm in patients with severe COPD. *Am J Respir Crit Care Med* 1997; 155: 1335–1340.
- 119 Gandevia SC, Leeper JB, McKenzie DK, *et al.* Discharge frequencies of parasternal intercostal and scalene motor units during breathing in normal and COPD subjects. *Am J Respir Crit Care Med* 1996; 153: 622–628.
- 120 Qin YY, Li RF, Wu GF, *et al.* Effect of tiotropium on neural respiratory drive during exercise in severe COPD. *Pulm Pharmacol Ther* 2015; 30: 51–56.
- 121 Yokoba M, Ichikawa T, Takakura A, *et al.* Aminophylline increases respiratory muscle activity during hypercapnia in humans. *Pulm Pharmacol Ther* 2015; 30: 96–101.
- 122 Gorman RB, McKenzie DK, Butler JE, *et al.* Diaphragm length and neural drive after lung volume reduction surgery. *Am J Respir Crit Care Med* 2005; 172: 1259–1266.
- 123 Firestone KS, Beck J, Stein H. Neurally adjusted ventilatory assist for noninvasive support in neonates. *Clin Perinatol* 2016; 43: 707–724.
- 124 Doorduyn J, Sinderby CA, Beck J, *et al.* Assisted ventilation in patients with acute respiratory distress syndrome: lung-distending pressure and patient-ventilator interaction. *Anesthesiology* 2015; 123: 181–190.
- 125 Sinderby C, Navalesi P, Beck J, *et al.* Neural control of mechanical ventilation in respiratory failure. *Nat Med* 1999; 5: 1433–1436.
- 126 Bellani G, Mauri T, Coppadoro A, *et al.* Estimation of patient's inspiratory effort from the electrical activity of the diaphragm. *Crit Care Med* 2013; 41: 1483–1491.
- 127 Dres M, Schmidt M, Ferre A, *et al.* Diaphragm electromyographic activity as a predictor of weaning failure. *Intensive Care Med* 2012; 38: 2017–2025.
- 128 Raux M, Straus C, Redolfi S, *et al.* Electroencephalographic evidence for pre-motor cortex activation during inspiratory loading in humans. *J Physiol (Lond)* 2007; 578: 569–578.
- 129 Jutand L, Tremoureux L, Pichon A, *et al.* Ventilatory response to exercise does not evidence electroencephalographical respiratory-related activation of the cortical premotor circuitry in healthy humans. *Acta Physiol* 2012; 205: 356–362.
- 130 McKay LC, Adams L, Frackowiak RS, *et al.* A bilateral cortico-bulbar network associated with breath holding in humans, determined by functional magnetic resonance imaging. *Neuroimage* 2008; 40: 1824–1832.
- 131 Hudson AL, Navarro-Sune X, Martinerie J, *et al.* Electroencephalographic detection of respiratory-related cortical activity in humans: from event-related approaches to continuous connectivity evaluation. *J Neurophysiol* 2016; 115: 2214–2223.
- 132 Tremoureux L, Raux M, Ranohavimparany A, *et al.* Electroencephalographic evidence for a respiratory-related cortical activity specific of the preparation of prephonatory breaths. *Respir Physiol Neurobiol* 2014; 204: 64–70.
- 133 Laviolette L, Nierat MC, Hudson AL, *et al.* The supplementary motor area exerts a tonic excitatory influence on corticospinal projections to phrenic motoneurons in awake humans. *PLoS One* 2013; 8: e62258.
- 134 Dubois M, Chenivresse C, Raux M, *et al.* Neurophysiological evidence for a cortical contribution to the wakefulness-related drive to breathe explaining hypocapnia-resistant ventilation in humans. *J Neurosci* 2016; 36: 10673–10682.
- 135 Tremoureux L, Raux M, Jutand L, *et al.* Sustained preinspiratory cortical potentials during prolonged inspiratory threshold loading in humans. *J Appl Physiol* 2010; 108: 1127–1133.
- 136 Raux M, Tyvaert L, Ferreira M, *et al.* Functional magnetic resonance imaging suggests automatization of the cortical response to inspiratory threshold loading in humans. *Respir Physiol Neurobiol* 2013; 189: 571–580.
- 137 Raux M, Ray P, Prella M, *et al.* Cerebral cortex activation during experimentally induced ventilator fighting in normal humans receiving noninvasive mechanical ventilation. *Anesthesiology* 2007; 107: 746–755.

- 138 Morawiec E, Raux M, Kindler F, *et al.* Expiratory load compensation is associated with electroencephalographic premotor potentials in humans. *J Appl Physiol* 2015; 118: 1023–1030.
- 139 Tremoureux L, Raux M, Hudson AL, *et al.* Does the supplementary motor area keep patients with Ondine's curse syndrome breathing while awake? *PLoS One* 2014; 9: e84534.
- 140 Sharman M, Gallea C, Lehongre K, *et al.* The cerebral cost of breathing: an FMRI case-study in congenital central hypoventilation syndrome. *PLoS one* 2014; 9: e107850.
- 141 Nierat MC, Demiri S, Dupuis-Lozeron E, *et al.* When breathing interferes with cognition: experimental inspiratory loading alters timed up-and-go test in normal humans. *PLoS One* 2016; 11: e0151625.
- 142 Launois C, Attali V, Georges M, *et al.* Cortical drive to breathe during wakefulness in patients with obstructive sleep apnea syndrome. *Sleep* 2015; 38: 1743–1749.
- 143 Georges M, Morawiec E, Raux M, *et al.* Cortical drive to breathe in amyotrophic lateral sclerosis: a dyspnoea-worsening defence? *Eur Respir J* 2016; 47: 1818–1828.
- 144 Navarro-Sune X, Hudson AL, De Vico Fallani F, *et al.* Riemannian geometry applied to detection of respiratory states from EEG signals: the basis for a brain-ventilator interface. *IEEE Trans Biomed Eng* 2017; 64: 1138–1148.
- 145 Hallett M. Transcranial magnetic stimulation and the human brain. *Nature* 2000; 406: 147–150.
- 146 Anand S, Hotson J. Transcranial magnetic stimulation: neurophysiological applications and safety. *Brain Cogn* 2002; 50: 366–386.
- 147 Keel JC, Smith MJ, Wassermann EM. A safety screening questionnaire for transcranial magnetic stimulation. *Clin Neurophysiol* 2001; 112: 720.
- 148 Demoule A, Verin E, Locher C, *et al.* Validation of surface recordings of the diaphragm response to transcranial magnetic stimulation in humans. *J Appl Physiol* 2003; 94: 453–461.
- 149 Straus C, Locher C, Zelter M, *et al.* Facilitation of the diaphragm response to transcranial magnetic stimulation by increases in human respiratory drive. *J Appl Physiol* 2004; 97: 902–912.
- 150 Mehiri S, Straus C, Arnulf I, *et al.* Responses of the diaphragm to transcranial magnetic stimulation during wake and sleep in humans. *Respir Physiol Neurobiol* 2006; 154: 406–418.
- 151 Demoule A, Verin E, Ross E, *et al.* Intracortical inhibition and facilitation of the response of the diaphragm to transcranial magnetic stimulation. *J Clin Neurophysiol* 2003; 20: 59–64.
- 152 Similowski T, Straus C, Coic L, *et al.* Facilitation-independent response of the diaphragm to cortical magnetic stimulation. *Am J Respir Crit Care Med* 1996; 154: 1771–1777.
- 153 Laguëny A, Arnaud A, Le Masson G, *et al.* Study of central and peripheral conduction to the diaphragm in 22 patients with definite multiple sclerosis. *Electromyogr Clin Neurophysiol* 1998; 38: 333–342.
- 154 Similowski T, Attali V, Bensimon G, *et al.* Diaphragmatic dysfunction and dyspnoea in amyotrophic lateral sclerosis. *Eur Respir J* 2000; 15: 332–337.
- 155 Wang W, Kang J, Kong D. The central motor conductivity of genioglossus in obstructive sleep apnoea. *Respirology* 2010; 15: 1209–1214.
- 156 Series F, Wang W, Similowski T. Corticomotor control of the genioglossus in awake OSAS patients: a transcranial magnetic stimulation study. *Respir Res* 2009; 10: 74.
- 157 Melo-Silva CA, Borel JC, Gakwaya S, *et al.* Acute upper airway muscle and inspiratory flow responses to transcranial magnetic stimulation during sleep in apnoeic patients. *Exp Physiol* 2013; 98: 946–956.
- 158 Melo-Silva CA, Gakwaya S, Rousseau E, *et al.* Consecutive transcranial magnetic stimulation twitches reduce flow limitation during sleep in apnoeic patients. *Exp Physiol* 2013; 98: 1366–1375.
- 159 Rousseau E, Gakwaya S, Melo-Silva CA, *et al.* Mechanical effects of repetitive transcranial magnetic stimulation of upper airway muscles in awake obstructive sleep apnoea subjects. *Exp Physiol* 2015; 100: 566–576.
- 160 Rousseau E, Melo-Silva CA, Gakwaya S, *et al.* Effects of repetitive transcranial magnetic stimulation of upper airway muscles during sleep in obstructive sleep apnea patients. *J Appl Physiol* 2016; 121: 1217–1225.
- 161 Hopkinson NS, Sharshar T, Dayer MJ, *et al.* The effect of acute non-invasive ventilation on corticospinal pathways to the respiratory muscles in chronic obstructive pulmonary disease. *Respir Physiol Neurobiol* 2012; 183: 41–47.
- 162 Hopkinson NS, Sharshar T, Ross ET, *et al.* Corticospinal control of respiratory muscles in chronic obstructive pulmonary disease. *Respir Physiol Neurobiol* 2004; 141: 1–12.
- 163 Duguet A, Demoule A, Gonzalez J, *et al.* Predicting the recovery of ventilatory activity in central respiratory paralysis. *Neurology* 2006; 67: 288–292.
- 164 Harraf F, Ward K, Man W, *et al.* Transcranial magnetic stimulation study of expiratory muscle weakness in acute ischemic stroke. *Neurology* 2008; 71: 2000–2007.
- 165 Houston JG, Fleet M, Cowan MD, *et al.* Comparison of ultrasound with fluoroscopy in the assessment of suspected hemidiaphragmatic movement abnormality. *Clin Radiol* 1995; 50: 95–98.
- 166 Matamis D, Soilemezi E, Tsagourias M, *et al.* Sonographic evaluation of the diaphragm in critically ill patients. Technique and clinical applications. *Intensive Care Med* 2013; 39: 801–810.
- 167 Boon AJ, Harper CJ, Ghahfarokhi LS, *et al.* Two-dimensional ultrasound imaging of the diaphragm: quantitative values in normal subjects. *Muscle Nerve* 2013; 47: 884–889.
- 168 Wait JL, Nahormek PA, Yost WT, *et al.* Diaphragmatic thickness-lung volume relationship *in vivo*. *J Appl Physiol* 1989; 67: 1560–1568.
- 169 Baldwin CE, Paratz JD, Bersten AD. Diaphragm and peripheral muscle thickness on ultrasound: intra-rater reliability and variability of a methodology using non-standard recumbent positions. *Respirology* 2011; 16: 1136–1143.
- 170 Cohn D, Benditt JO, Eveloff S, *et al.* Diaphragm thickening during inspiration. *J Appl Physiol* 1997; 83: 291–296.
- 171 Enright S, Chatham K, Ionescu AA, *et al.* The influence of body composition on respiratory muscle, lung function and diaphragm thickness in adults with cystic fibrosis. *J Cyst Fibros* 2007; 6: 384–390.
- 172 Baria MR, Shahgholi L, Sorenson EJ, *et al.* B-mode ultrasound assessment of diaphragm structure and function in patients with COPD. *Chest* 2014; 146: 680–685.
- 173 Hellyer NJ, Andreas NM, Bernstetter AS, *et al.* Comparison of diaphragm thickness measurements among postures *via* ultrasound imaging. *PM R* 2017; 9: 21–25.
- 174 McCool FD, Benditt JO, Conomos P, *et al.* Variability of diaphragm structure among healthy individuals. *Am J Respir Crit Care Med* 1997; 155: 1323–1328.

- 175 Dufresne V, Knoop C, Van Muylem A, *et al.* Effect of systemic inflammation on inspiratory and limb muscle strength and bulk in cystic fibrosis. *Am J Respir Crit Care Med* 2009; 180: 153–158.
- 176 Summerhill EM, El-Sameed YA, Glidden TJ, *et al.* Monitoring recovery from diaphragm paralysis with ultrasound. *Chest* 2008; 133: 737–743.
- 177 Gottesman E, McCool FD. Ultrasound evaluation of the paralyzed diaphragm. *Am J Respir Crit Care Med* 1997; 155: 1570–1574.
- 178 Bruin PFD, Ueki J, Bush A, *et al.* Diaphragm thickness and inspiratory strength in patients with Duchenne muscular dystrophy. *Thorax* 1997; 52: 474–475.
- 179 Goligher EC, Laghi F, Detsky ME, *et al.* Measuring diaphragm thickness with ultrasound in mechanically ventilated patients: feasibility, reproducibility and validity. *Intensive Care Med* 2015; 41: 642–649.
- 180 Vivier E, Dessap AM, Dimassi S, *et al.* Diaphragm ultrasonography to estimate the work of breathing during non-invasive ventilation. *Intensive Care Med* 2012; 38: 796–803.
- 181 Dube BP, Dres M, Mayaux J, *et al.* Ultrasound evaluation of diaphragm function in mechanically ventilated patients: comparison to phrenic stimulation and prognostic implications. *Thorax* 2017; 72: 811–818.
- 182 DiNino E, Gartman EJ, Sethi JM, *et al.* Diaphragm ultrasound as a predictor of successful extubation from mechanical ventilation. *Thorax* 2014; 69: 423–427.
- 183 Ferrari G, De Filippi G, Elia F, *et al.* Diaphragm ultrasound as a new index of discontinuation from mechanical ventilation. *Crit Ultrasound J* 2014; 6: 8.
- 184 Dube BP, Dres M, Mayaux J, *et al.* Ultrasound evaluation of diaphragm function in mechanically ventilated patients: comparison to phrenic stimulation and prognostic implications. *Thorax* 2017; 72: 811–818.
- 185 Goligher EC, Fan E, Herridge MS, *et al.* Evolution of diaphragm thickness during mechanical ventilation: impact of inspiratory effort. *Am J Respir Crit Care Med* 2015; 192: 1080–1088.
- 186 Grosu HB, Lee YI, Lee J, *et al.* Diaphragm muscle thinning in patients who are mechanically ventilated. *Chest* 2012; 142: 1455–1460.
- 187 Goligher EC, Laghi F, Detsky ME, *et al.* Measuring diaphragm thickness with ultrasound in mechanically ventilated patients: feasibility, reproducibility and validity. *Intensive Care Med* 2015; 41: 642–649.
- 188 Umbrello M, Formenti P, Longhi D, *et al.* Diaphragm ultrasound as indicator of respiratory effort in critically ill patients undergoing assisted mechanical ventilation: a pilot clinical study. *Crit Care* 2015; 19: 161.
- 189 Dres M, Dube BP, Mayaux J, *et al.* Coexistence and impact of limb muscle and diaphragm weakness at time of liberation from mechanical ventilation in medical intensive care unit patients. *Am J Respir Crit Care Med* 2017; 195: 57–66.
- 190 Antenora F, Fantini R, Iattoni A, *et al.* Prevalence and outcomes of diaphragmatic dysfunction assessed by ultrasound technology during acute exacerbation of COPD: a pilot study. *Respirology* 2017; 22: 338–344.
- 191 Boussuges A, Gole Y, Blanc P. Diaphragmatic motion studied by M-mode ultrasonography: methods, reproducibility, and normal values. *Chest* 2009; 135: 391–400.
- 192 Kim WY, Suh HJ, Hong SB, *et al.* Diaphragm dysfunction assessed by ultrasonography: influence on weaning from mechanical ventilation. *Crit Care Med* 2011; 39: 2627–2630.
- 193 Orde SR, Boon AJ, Firth DG, *et al.* Use of angle-independent M-mode sonography for assessment of diaphragm displacement. *J Ultrasound Med* 2016; 35: 2615–2621.
- 194 Jones AYMP, Ngai SPCP, Ying MTCP, *et al.* Sonographic evaluation of diaphragmatic function during breathing control. *Physiother Theory Pract* 2017; 33: 560–567.
- 195 Kim SH, Na S, Choi JS, *et al.* An evaluation of diaphragmatic movement by M-mode sonography as a predictor of pulmonary dysfunction after upper abdominal surgery. *Anesth Analg* 2010; 110: 1349–1354.
- 196 Numis FG, Morelli L, Bosso G, *et al.* Diaphragmatic motility assessment in COPD exacerbation, early detection of non-invasive mechanical ventilation failure: a pilot study. *Crit Ultrasound J* 2014; 6: Suppl. 2, A6.
- 197 Houston JG, Morris AD, Grosset DG, *et al.* Ultrasonic evaluation of movement of the diaphragm after acute cerebral infarction. *J Neurol Neurosurg Psychiatry* 1995; 58: 738–741.
- 198 Lerolle N, Guerot E, Dimassi S, *et al.* Ultrasonographic diagnostic criterion for severe diaphragmatic dysfunction after cardiac surgery. *Chest* 2009; 135: 401–407.
- 199 Spadaro S, Grasso S, Mauri T, *et al.* Can diaphragmatic ultrasonography performed during the T-tube trial predict weaning failure? The role of diaphragmatic rapid shallow breathing index. *Crit Care* 2016; 20: 305.
- 200 Trinavarat P, Riccabona M. Potential of ultrasound in the pediatric chest. *Eur J Radiol* 2014; 83: 1507–1518.
- 201 Cala SJ, Kenyon CM, Ferrigno G, *et al.* Chest wall and lung volume estimation by optical reflectance motion analysis. *J Appl Physiol* 1996; 81: 2680–2689.
- 202 Aliverti A, Dellaca R, Pelosi P, *et al.* Compartmental analysis of breathing in the supine and prone positions by optoelectronic plethysmography. *Ann Biomed Eng* 2001; 29: 60–70.
- 203 Vogiatzis I, Georgiadou O, Golemati S, *et al.* Patterns of dynamic hyperinflation during exercise and recovery in patients with severe chronic obstructive pulmonary disease. *Thorax* 2005; 60: 723–729.
- 204 Aliverti A, Stevenson N, Dellaca RL, *et al.* Regional chest wall volumes during exercise in chronic obstructive pulmonary disease. *Thorax* 2004; 59: 210–216.
- 205 Georgiadou O, Vogiatzis I, Stratakos G, *et al.* Effects of rehabilitation on chest wall volume regulation during exercise in COPD patients. *Eur Respir J* 2007; 29: 284–291.
- 206 Wilkens H, Weingard B, Lo Mauro A, *et al.* Breathing pattern and chest wall volumes during exercise in patients with cystic fibrosis, pulmonary fibrosis and COPD before and after lung transplantation. *Thorax* 2010; 65: 808–814.
- 207 Mauro A L, D'Angelo MG, Romei M, *et al.* Abdominal volume contribution to tidal volume as an early indicator of respiratory impairment in Duchenne muscular dystrophy. *Eur Respir J* 2010; 35: 1118–1125.
- 208 LoMauro A, Romei M, D'Angelo MG, *et al.* Determinants of cough efficiency in Duchenne muscular dystrophy. *Pediatr Pulmonol* 2014; 49: 357–365.
- 209 Romei M, D'Angelo MG, LoMauro A, *et al.* Low abdominal contribution to breathing as daytime predictor of nocturnal desaturation in adolescents and young adults with Duchenne muscular dystrophy. *Respir Med* 2012; 106: 276–283.
- 210 D'Angelo MG, Romei M, Lo Mauro A, *et al.* Respiratory pattern in an adult population of dystrophic patients. *J Neurol Sci* 2011; 306: 54–61.

- 211 Layton AM, Moran SL, Roychoudhury A, *et al.* Non-invasive measurement of abnormal ventilatory mechanics in amyotrophic lateral sclerosis. *Muscle Nerve* 2016; 54: 270–276.
- 212 Lunardi AC, Paisani Dde M, Tanaka C, *et al.* Impact of laparoscopic surgery on thoracoabdominal mechanics and inspiratory muscular activity. *Respir Physiol Neurobiol* 2013; 186: 40–44.
- 213 Acosta J, Bradley A, Raja V, *et al.* Exercise improvement after pectus excavatum repair is not related to chest wall function. *Eur J Cardiothorac Surg* 2014; 45: 544–548.
- 214 Elshafie G, Acosta J, Aliverti A, *et al.* Chest wall mechanics before and after diaphragm plication. *J Cardiothorac Surg* 2016; 11: 25.
- 215 Laviola M, Zanini A, Priori R, *et al.* Thoraco-abdominal asymmetry and asynchrony in congenital diaphragmatic hernia. *Pediatr Pulmonol* 2015; 50: 915–924.
- 216 Sarmiento A, de Andrade AF, Lima IN, *et al.* Air stacking: a detailed look into physiological acute effects on cough peak flow and chest wall volumes of healthy subjects. *Respir Care* 2017; 62: 432–443.
- 217 Jde M B, Aliverti A, Rattes C, *et al.* The expansion of the pulmonary rib cage during breath stacking is influenced by age in obese women. *PLoS One* 2014; 9: e110959.
- 218 de Sa RB, Pessoa MF, Cavalcanti AGL, *et al.* Immediate effects of respiratory muscle stretching on chest wall kinematics and electromyography in COPD patients. *Respir Physiol Neurobiol* 2017; 242: 1–7.
- 219 Paisani DM, Lunardi AC, da Silva CC, *et al.* Volume rather than flow incentive spirometry is effective in improving chest wall expansion and abdominal displacement using optoelectronic plethysmography. *Respir Care* 2013; 58: 1360–1366.
- 220 Brandao DC, Lage SM, Britto RR, *et al.* Chest wall regional volume in heart failure patients during inspiratory loaded breathing. *Respir Physiol Neurobiol* 2012; 180: 269–274.
- 221 Albuquerque AL, Quaranta M, Chakrabarti B, *et al.* Exercise performance and differences in physiological response to pulmonary rehabilitation in severe chronic obstructive pulmonary disease with hyperinflation. *J Bras Pneumol* 2016; 42: 121–129.
- 222 Dellaca RL, Ventura ML, Zannin E, *et al.* Measurement of total and compartmental lung volume changes in newborns by optoelectronic plethysmography. *Pediatr Res* 2010; 67: 11–16.
- 223 LoMauro A, Aliverti A, Mastella C, *et al.* Spontaneous breathing pattern as respiratory functional outcome in children with spinal muscular atrophy (SMA). *PloS One* 2016; 11: e0165818.
- 224 Harlaar L, Ciet P, van der Ploeg A, *et al.* Imaging of respiratory muscles in neuromuscular disease: a review. *Neuromuscul Disord* 2018; 28: 246–256.
- 225 Mogalle K, Perez-Rovira A, Ciet P, *et al.* Quantification of diaphragm mechanics in Pompe disease using dynamic 3D MRI. *PloS One* 2016; 11: e0158912.
- 226 Nierat MC, Dube BP, Llontop C, *et al.* Measuring ventilatory activity with structured light plethysmography (SLP) reduces instrumental observer effect and preserves tidal breathing variability in healthy and COPD. *Front Physiol* 2017; 8: 316.
- 227 Motamedi-Fakhr S, Iles R, Barney A, *et al.* Evaluation of the agreement of tidal breathing parameters measured simultaneously using pneumotachography and structured light plethysmography. *Physiol Rep* 2017; 5: e13124.
- 228 Motamedi-Fakhr S, Wilson RC, Iles R. Tidal breathing patterns derived from structured light plethysmography in COPD patients compared with healthy subjects. *Med Devices (Auckl)* 2017; 10: 1–9.
- 229 Elshafie G, Kumar P, Motamedi-Fakhr S, *et al.* Measuring changes in chest wall motion after lung resection using structured light plethysmography: a feasibility study. *Interact Cardiovasc Thorac Surg* 2016; 23: 544–547.
- 230 Hmeidi H, Motamedi-Fakhr S, Chadwick E, *et al.* Tidal breathing parameters measured using structured light plethysmography in healthy children and those with asthma before and after bronchodilator. *Physiol Rep* 2017; 5: e13168.
- 231 Guenette JA, Vogiatzis I, Zakynthinos S, *et al.* Human respiratory muscle blood flow measured by near-infrared spectroscopy and indocyanine green. *J Appl Physiol* 2008; 104: 1202–1210.
- 232 Vogiatzis I, Athanasopoulos D, Habazettl H, *et al.* Intercostal muscle blood flow limitation in athletes during maximal exercise. *J Physiol* 2009; 587: 3665–3677.
- 233 Vogiatzis I, Athanasopoulos D, Habazettl H, *et al.* Intercostal muscle blood flow limitation during exercise in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2010; 182: 1105–1113.
- 234 Habazettl H, Athanasopoulos D, Kuebler WM, *et al.* Near-infrared spectroscopy and indocyanine green derived blood flow index for noninvasive measurement of muscle perfusion during exercise. *J Appl Physiol* 2010; 108: 962–967.
- 235 Guenette JA, Henderson WR, Dominelli PB, *et al.* Blood flow index using near-infrared spectroscopy and indocyanine green as a minimally invasive tool to assess respiratory muscle blood flow in humans. *Am J Physiol Regul Integr Comp Physiol* 2011; 300: R984–R992.
- 236 Dominelli PB, Render JN, Molgat-Seon Y, *et al.* Oxygen cost of exercise hyperpnoea is greater in women compared with men. *J Physiol* 2015; 593: 1965–1979.
- 237 Topin N, Mucci P, Hayot M, *et al.* Gender influence on the oxygen consumption on the respiratory muscles in young and older healthy individuals. *Int J Sports Med* 2003; 24: 559–564.
- 238 Kress JP, Pohlman AS, Alverdy J, *et al.* The impact of morbid obesity on oxygen cost of breathing ($\text{VO}_{2\text{RESP}}$) at rest. *Am J Respir Crit Care Med* 1999; 160: 883–886.
- 239 Sharp JT, Henry JP, Sweany SK, *et al.* The total work of breathing in normal and obese men. *J Clin Invest* 1964; 43: 728–739.
- 240 Weyland W, Schuhmann M, Rathgeber J, *et al.* Oxygen cost of breathing for assisted spontaneous breathing modes: investigation into three states of pulmonary function. *Intensive Care Med* 1995; 21: 211–217.
- 241 Schols AM, Soeters PB, Mostert R, *et al.* Energy balance in chronic obstructive pulmonary disease. *Am Rev Respir Dis* 1991; 143: 1248–1252.
- 242 Bell SC, Saunders MJ, Elborn JS, *et al.* Resting energy expenditure and oxygen cost of breathing in patients with cystic fibrosis. *Thorax* 1996; 51: 126–131.
- 243 Katsardis CV, Desmond KJ, Coates AL. Measuring the oxygen cost of breathing in normal adults and patients with cystic fibrosis. *Respir Physiol* 1986; 65: 257–266.
- 244 Thomas J, Enecio CE, Chehreh MN, *et al.* Oxygen cost of breathing III: studies in asthmatic children. *J Natl Med Assoc* 1976; 68: 374–377.

- 245 Harden KA, Young RC Jr, Carr C, *et al.* Oxygen cost of breathing. II. Studies in patients with sarcoidosis. *Am Rev Respir Dis* 1968; 97: 1127–1130.
- 246 Kurotobi T, Sato H, Yokoyama H, *et al.* Respiratory oxygen cost for dead space challenge is characteristically increased during exercise in patients with chronic heart failure: does it further decrease exercise capacity? *J Card Fail* 1997; 3: 181–188.
- 247 Reddy HK, McElroy PA, Janicki JS, *et al.* Response in oxygen uptake and ventilation during stair climbing in patients with chronic heart failure. *Am J Cardiol* 1989; 63: 222–225.
- 248 Alexiou S, Panitch HB. Physiology of non-invasive respiratory support. *Semin Fetal Neonatal Med* 2016; 21: 174–180.
- 249 Duiverman ML, Arellano-Maric MP, Windisch W. Long-term noninvasive ventilation in patients with chronic hypercapnic respiratory failure: assisting the diaphragm, but threatening the heart? *Curr Opin Pulm Med* 2016; 22: 130–137.
- 250 White DP, Criner GJ, Dreher M, *et al.* The role of noninvasive ventilation in the management and mitigation of exacerbations and hospital admissions/readmissions for the patient with moderate to severe COPD (multimedia activity). *Chest* 2015; 147: 1704–1705.
- 251 Lee CC, Mankodi D, Shaharyar S, *et al.* High flow nasal cannula versus conventional oxygen therapy and non-invasive ventilation in adults with acute hypoxemic respiratory failure: a systematic review. *Respir Med* 2016; 121: 100–108.
- 252 Joliet P, Ouanes-Besbes L, Abroug F, *et al.* A multicenter randomized trial assessing the efficacy of helium/oxygen in severe exacerbations of chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2017; 195: 871–880.
- 253 Bieli C, Summermatter S, Boutellier U, *et al.* Respiratory muscle training improves respiratory muscle endurance but not exercise tolerance in children with cystic fibrosis. *Pediatr Pulmonol* 2017; 52: 331–336.
- 254 Dellweg D, Reissig K, Hoehn E, *et al.* Inspiratory muscle training during rehabilitation in successfully weaned hypercapnic patients with COPD. *Respir Med* 2017; 123: 116–123.
- 255 Spruit MA, Singh SJ, Garvey C, *et al.* An official American Thoracic Society/European Respiratory Society statement: key concepts and advances in pulmonary rehabilitation. *Am J Respir Crit Care Med* 2013; 188: e13–e64.
- 256 Barreiro E, de la Puente B, Minguella J, *et al.* Oxidative stress and respiratory muscle dysfunction in severe chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2005; 171: 1116–1124.
- 257 Barreiro E, Ferrer D, Sanchez F, *et al.* Inflammatory cells and apoptosis in respiratory and limb muscles of patients with COPD. *J Appl Physiol* 2011; 111: 808–817.
- 258 Doucet M, Degibare R, Joannisse DR, *et al.* Adaptation of the diaphragm and the vastus lateralis in mild-to-moderate COPD. *Eur Respir J* 2004; 24: 971–979.
- 259 Marin-Corral J, Minguella J, Ramirez-Sarmiento AL, *et al.* Oxidised proteins and superoxide anion production in the diaphragm of severe COPD patients. *Eur Respir J* 2009; 33: 1309–1319.
- 260 Rabinovich RA, Bastos R, Ardite E, *et al.* Mitochondrial dysfunction in COPD patients with low body mass index. *Eur Respir J* 2007; 29: 643–650.
- 261 Barreiro E, Coronell C, Lavina B, *et al.* Aging, sex differences, and oxidative stress in human respiratory and limb muscles. *Free Radic Biol Med* 2006; 41: 797–809.
- 262 Barreiro E, Nowinski A, Gea J, *et al.* Oxidative stress in the external intercostal muscles of patients with obstructive sleep apnoea. *Thorax* 2007; 62: 1095–1101.
- 263 Pascual-Guardia S, Arbol F, Sanchez E, *et al.* [Inflammation and oxidative stress in respiratory and limb muscles of patients with severe sepsis]. *Med Clin* 2013; 141: 194–200.
- 264 Pascual-Guardia S, Wodja E, Gorostiza A, *et al.* [Improvement in quality of life and exercise capacity without muscular biology changes after general training in patients with severe chronic obstructive pulmonary disease]. *Med Clin* 2013; 140: 200–206.
- 265 Pasto M, Gea J, Blanco M, *et al.* [Metabolic activity of the external intercostal muscle of patients with COPD]. *Arch Bronconeumol* 2001; 37: 108–114.
- 266 Maltais F, Decramer M, Casaburi R, *et al.* An official American Thoracic Society/European Respiratory Society statement: update on limb muscle dysfunction in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2014; 189: e15–e62.
- 267 Levine S, Kaiser L, Leforovich J, *et al.* Cellular adaptations in the diaphragm in chronic obstructive pulmonary disease. *N Engl J Med* 1997; 337: 1799–1806.
- 268 Levine S, Gregory C, Nguyen T, *et al.* Bioenergetic adaptation of individual human diaphragmatic myofibers to severe COPD. *J Appl Physiol* 2002; 92: 1205–1213.
- 269 Gea J, Hamid Q, Czaika G, *et al.* Expression of myosin heavy-chain isoforms in the respiratory muscles following inspiratory resistive breathing. *Am J Respir Crit Care Med* 2000; 161: 1274–1278.
- 270 Zhu E, Petrof BJ, Gea J, *et al.* Diaphragm muscle fiber injury after inspiratory resistive breathing. *Am J Respir Crit Care Med* 1997; 155: 1110–1116.
- 271 Orozco-Levi M, Gea J, Lloreta JL, *et al.* Subcellular adaptation of the human diaphragm in chronic obstructive pulmonary disease. *Eur Respir J* 1999; 13: 371–378.
- 272 Orozco-Levi M, Lloreta J, Minguella J, *et al.* Injury of the human diaphragm associated with exertion and chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2001; 164: 1734–1739.
- 273 Engelen MP, Orozco-Levi M, Deutz NE, *et al.* Glutathione and glutamate levels in the diaphragm of patients with chronic obstructive pulmonary disease. *Eur Respir J* 2004; 23: 545–551.
- 274 Sanchez J, Derenne JP, Debesse B, *et al.* Typology of the respiratory muscles in normal men and in patients with moderate chronic respiratory diseases. *Bull Eur Physiopathol Respir* 1982; 18: 901–914.
- 275 Levine S, Nguyen T, Friscia M, *et al.* Parasternal intercostal muscle remodeling in severe chronic obstructive pulmonary disease. *J Appl Physiol* 2006; 101: 1297–1302.
- 276 Jimenez-Fuentes MA, Gea J, Aguar MC, *et al.* [Capillary density and respiratory function in the external intercostal muscle]. *Arch Bronconeumol* 1999; 35: 471–476.
- 277 Ju S, Lee SJ, Park MJ, *et al.* Clinical importance of cross-sectional area of intercostal muscles in patients with chronic obstructive pulmonary disease. *Clin Respir J* 2018; 12: 939–947.

- 278 Ramirez-Sarmiento A, Orozco-Levi M, Guell R, *et al.* Inspiratory muscle training in patients with chronic obstructive pulmonary disease: structural adaptation and physiologic outcomes. *Am J Respir Crit Care Med* 2002; 166: 1491–1497.
- 279 Jaber S, Petrof BJ, Jung B, *et al.* Rapidly progressive diaphragmatic weakness and injury during mechanical ventilation in humans. *Am J Respir Crit Care Med* 2011; 183: 364–371.
- 280 Levine S, Nguyen T, Taylor N, *et al.* Rapid disuse atrophy of diaphragm fibers in mechanically ventilated humans. *N Engl J Med* 2008; 358: 1327–1335.
- 281 Garcia-Cazarin ML, Gamboa JL, Andrade FH. Rat diaphragm mitochondria have lower intrinsic respiratory rates than mitochondria in limb muscles. *Am J Physiol Regul Integr Comp Physiol* 2011; 300: R1311–R1315.
- 282 Gamboa JL, Andrade FH. Mitochondrial content and distribution changes specific to mouse diaphragm after chronic normobaric hypoxia. *Am J Physiol Regul Integr Comp Physiol* 2010; 298: R575–R583.
- 283 Picard M, Godin R, Sinnreich M, *et al.* The mitochondrial phenotype of peripheral muscle in chronic obstructive pulmonary disease: disuse or dysfunction? *Am J Respir Crit Care Med* 2008; 178: 1040–1047.
- 284 Puente-Maestu L, Perez-Parra J, Godoy R, *et al.* Abnormal mitochondrial function in locomotor and respiratory muscles of COPD patients. *Eur Respir J* 2009; 33: 1045–1052.
- 285 Wijnhoven JH, Janssen AJ, van Kuppevelt TH, *et al.* Metabolic capacity of the diaphragm in patients with COPD. *Respir Med* 2006; 100: 1064–1071.
- 286 Ribera F, N'Guessan B, Zoll J, *et al.* Mitochondrial electron transport chain function is enhanced in inspiratory muscles of patients with chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2003; 167: 873–879.
- 287 Fredriksson K, Rooyackers O. Mitochondrial function in sepsis: respiratory *versus* leg muscle. *Crit Care Med* 2007; 35: Suppl. 9, S449–S453.
- 288 Bernard N, Matecki S, Py G, *et al.* Effects of prolonged mechanical ventilation on respiratory muscle ultrastructure and mitochondrial respiration in rabbits. *Intensive Care Med* 2003; 29: 111–118.
- 289 Wijnhoven HJ, Ennen L, Rodenburg RJ, *et al.* Mitochondrial function in diaphragm of emphysematous hamsters after treatment with nandrolone. *Int J Chron Obstruct Pulmon Dis* 2006; 1: 83–89.
- 290 Powers SK, Criswell D. Adaptive strategies of respiratory muscles in response to endurance exercise. *Med Sci Sports Exerc* 1996; 28: 1115–1122.
- 291 Ottenheijm CA, Heunks LM, Li YP, *et al.* Activation of the ubiquitin-proteasome pathway in the diaphragm in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2006; 174: 997–1002.
- 292 Ottenheijm CA, Heunks LM, Sieck GC, *et al.* Diaphragm dysfunction in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2005; 172: 200–205.
- 293 Hussain SN, Mofarrah M, Sigala I, *et al.* Mechanical ventilation-induced diaphragm disuse in humans triggers autophagy. *Am J Respir Crit Care Med* 2010; 182: 1377–1386.
- 294 Picard M, Jung B, Liang F, *et al.* Mitochondrial dysfunction and lipid accumulation in the human diaphragm during mechanical ventilation. *Am J Respir Crit Care Med* 2012; 186: 1140–1149.
- 295 Jaitovich A, Barreiro E. Skeletal muscle dysfunction in chronic obstructive pulmonary disease. What we know and can do for our patients. *Am J Respir Crit Care Med* 2018; 198: 175–186.
- 296 Casadevall C, Coronell C, Ramirez-Sarmiento AL, *et al.* Upregulation of pro-inflammatory cytokines in the intercostal muscles of COPD patients. *Eur Respir J* 2007; 30: 701–707.
- 297 Volianitis S, McConnell AK, Jones DA. Assessment of maximum inspiratory pressure. Prior submaximal respiratory muscle activity ('warm-up') enhances maximum inspiratory activity and attenuates the learning effect of repeated measurement. *Respiration* 2001; 68: 22–27.
- 298 Fiz JA, Montserrat JM, Picado C, *et al.* How many maneuvers should be done to measure maximal inspiratory mouth pressure in patients with chronic air-flow obstruction. *Thorax* 1989; 44: 419–421.
- 299 Lofaso F, Nicot F, Lejaille M, *et al.* Sniff nasal inspiratory pressure: what is the optimal number of sniffs? *Eur Respir J* 2006; 27: 980–982.
- 300 Terzi N, Corne F, Mouadil A, *et al.* Mouth and nasal inspiratory pressure: learning effect and reproducibility in healthy adults. *Respiration* 2010; 80: 379–386.
- 301 Bianchi C, Baiardi P. Cough peak flows: standard values for children and adolescents. *Am J Phys Med Rehab* 2008; 87: 461–467.
- 302 Bach JR, Saporito LR. Criteria for extubation and tracheostomy tube removal for patients with ventilatory failure – a different approach to weaning. *Chest* 1996; 110: 1566–1571.
- 303 Miller JM, Moxham J, Green M. The maximal sniff in the assessment of diaphragm function in man. *Clin Sci* 1985; 69: 91–96.
- 304 Polkey MI, Harris ML, Hughes PD, *et al.* The contractile properties of the elderly human diaphragm. *Am J Respir Crit Care Med* 1997; 155: 1560–1564.
- 305 Hughes PD, Polkey MI, Harris ML, *et al.* Diaphragm strength in chronic heart failure. *Am J Respir Crit Care Med* 1999; 160: 529–534.
- 306 Kabitz HJ, Schwoerer A, Bremer HC, *et al.* Impairment of respiratory muscle function in pulmonary hypertension. *Clin Sci* 2008; 114: 165–171.
- 307 Hug F, Raux M, Prella M, *et al.* Optimized analysis of surface electromyograms of the scalenes during quiet breathing in humans. *Respir Physiol Neurobiol* 2006; 150: 75–81.
- 308 Chuang SY, Teng A, Butler JE, *et al.* Validation of a quantitative method to measure neural respiratory drive in children during sleep. *Respir Physiol Neurobiol* 2017; 239: 75–80.
- 309 Allen GM, McKenzie DK, Gandevia SC, *et al.* Reduced voluntary drive to breathe in asthmatic subjects. *Respir Physiol* 1993; 93: 29–40.
- 310 Hodges PW, Heijnen I, Gandevia SC. Postural activity of the diaphragm is reduced in humans when respiratory demand increases. *J Physiol (Lond)* 2001; 537: 999–1008.
- 311 Hodges PW, Gandevia SC. Pitfalls of intramuscular electromyographic recordings from the human costal diaphragm. *Clin Neurophysiol* 2000; 111: 1420–1424.
- 312 Luo YM, Polkey MI, Johnson LC, *et al.* Diaphragm EMG measured by cervical magnetic and electrical phrenic nerve stimulation. *J Appl Physiol* 1998; 85: 2089–2099.
- 313 McKenzie DK, Gandevia SC. Phrenic nerve conduction times and twitch pressures of the human diaphragm. *J Appl Physiol* 1985; 58: 1496–1504.

- 314 Chokroverty S, Hening W, Wright D, *et al.* Magnetic brain stimulation: safety studies. *Electroencephalogr Clin Neurophysiol* 1995; 97: 36–42.
- 315 Ngomo S, Leonard G, Moffet H, *et al.* Comparison of transcranial magnetic stimulation measures obtained at rest and under active conditions and their reliability. *J Neurosci Methods* 2012; 205: 65–71.
- 316 Malcolm MP, Triggs WJ, Light KE, *et al.* Reliability of motor cortex transcranial magnetic stimulation in four muscle representations. *Clin Neurophysiol* 2006; 117: 1037–1046.
- 317 Kimberley TJ, Borich MR, Prochaska KD, *et al.* Establishing the definition and inter-rater reliability of cortical silent period calculation in subjects with focal hand dystonia and healthy controls. *Neurosci Lett* 2009; 464: 84–87.
- 318 Flitman SS, Grafman J, Wassermann EM, *et al.* Linguistic processing during repetitive transcranial magnetic stimulation. *Neurology* 1998; 50: 175–181.