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Respiratory muscle activation and action during voluntary cough in healthy humans

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ABSTRACT

Cough is a defensive airway reflex consisting of a modified respiratory act which involves the sequential activation of several laryngeal and respiratory muscles. The contraction of the latter results in thoraco-abdominal volume variations in order to provide enough amount of air available, the operating volume (OV), to be expelled. Because both posture and OV could influence muscular activation and thoraco-abdominal displacements during voluntary cough, we aimed to verify if and how they play a role during inspiratory (ICP) and expiratory (ECP) cough phases, in terms of flow, volumes and surface electromyography activity (sEMG). In 10 healthy subjects, we measured sEMG of 7 muscles (scalene, sternocleidomastoid, parasternal, intercostal, diaphragm (assessed at the 8th intercostal space), external abdominal oblique and rectus abdominis) in supine and seated position during cough maneuvers performed at 4 different OV measured by opto-electronic plethismography: total lung capacity (TLC), functional residual capacity and two intermediate volumes. The amplitude of sEMG signals tended to be maximal at TLC ($p < 0.005$) during ICP in the neck and parasternal muscles and during ECP in abdominal muscles. Postures slightly affected only sEMG of the thoracic muscles. sEMG data were similar ($p > 0.05$) in the other OV, but cough peak flow increased with OV. Thoraco-abdominal volume variations during cough were unaffected by posture and OV as well, being predominantly thoracic (supine: 60 and 64%; seated: 68 and 69%, respectively during ICP and ECP).

Our results suggest that voluntary cough OV or posture do not have an important effect on voluntary cough that seems more likely to be resulting from a motor mechanism that activates a synergetic antagonistic contraction of inspiratory and expiratory muscles leading to a specific thoraco-abdominal pattern, in which the rib cage is the predominant.

1. Introduction

Cough is an important defence mechanism: its main function is to maintain airway clearance by removing mucus and/or foreign bodies from the lower respiratory tract. Cough is vagally mediated and it is the result of the coordinated activity of various respiratory muscles. Inspiratory muscles contract to reach the lung volume needed to generate the high-velocity of expiratory flow (Gandevia et al., 1990; Kobayashi et al., 1992). The contraction of the expiratory muscles against a closed glottis during the compressive phase builds up high positive intrapleural and intra-airway pressures to develop the adequate peak expiratory flow rates during the expulsive phase when glottis opens. The electrical activity of the muscles can be recorded using non-invasive surface transcutaneous electromyography (sEMG) to determine muscle capacity in terms of activation and contractile function (Hutten et al., 2010; Maarsingh et al., 2000; Duiverman et al., 2004). The electromyography of respiratory muscles has been studied on animals and humans mainly during cough induced by several nebulized agents. During cough, the diaphragm, the most important respiratory muscles, has been studied only with invasive procedures on animals, the intercostal muscles have been poorly studied (Fontana and Lavorini, 2006; Lasserson et al., 2006) whereas more information are available on the electrical activity of abdominal muscles in humans (Fontana and Lavorini, 2006; Fontana et al., 1997; Vovk et al., 2007; Cox et al., 1984; Lasserson et al., 2006). Taken together all these pieces of information, we learned that inspiratory muscles extend their electrical activity until the early stages of the expulsive cough phase (Kobayashi et al., 1992) while a certain degree of abdominal muscles activation is present in the late inspiratory phase. This suggests coactivation of antagonist muscles to control inspiratory volume of cough, flow and pressure generation as well as counteract the chest wall distortion secondary to the violent muscular activity during cough

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(Fontana and Lavorini, 2006).

The operating volume, *i.e.* the volume inspired at the end of the inspiratory cough phase, is the most important determinant of peak cough flow as it affects the expiratory muscle length and therefore their efficiency of contraction (Smith et al., 2012).

Because direct comparison of the respiratory muscles activation and action during cough has not been made, the purpose of this study was to combine measurements of sEMG and chest wall volumes variations in order to get a more comprehensive evaluation of the action of respiratory muscles during cough. We used opto-electronic plethysmography (Cala et al., 1996) because it provides not only total but also compartmental chest wall volume variations resulting from muscles contraction. In addition, the system was already proved to be compatible with cough manoeuvre not only in healthy subjects, but also in patients (Smith et al., 2012; LoMauro et al., 2014a, 2018; Cesareo et al., 2018).

We want to verify if muscular activity and therefore the thoraco-abdominal displacement would be influenced by the operating volumes. In addition, we explored whether posture could effect as well, because in the supine position the hydrostatic forces displace the diaphragm cranially, lung volume decreases, expiratory muscles lengthens and abdominal compliance reduces.

2. Methods

2.1. Subjects and ethics statement

This is a retrospective study of data that have been recorded between May and August 2012 on students, colleagues and collaborators working in our laboratory and therefore with experience in respiratory manoeuvres.

Exclusion criteria: respiratory, cardiovascular or neuromuscular diseases, prior chest wall surgery, spinal and ribcage deformities.

All the subjects signed a written informed consent form, as approved by the Local Ethical Committee of IRCCS “E. Medea” Institute according to the declaration of Helsinki.

2.2. Flow and volumes measurements

The flow at the mouth was measured with the subject wearing a nose clip and breathing through a pneumotacograph (Heated Pneumotach 800 L; Hans Rudolph®, Shawnee, USA) attached to a snorkel rubber mouthpiece. By definition, peak cough flow was the minimum value reached during the expiratory cough phase.

Synchronized measurements of rib cage, abdominal and chest wall volume were performed using Opto-Electronic Plethysmography (OEP System; BTS, Milan, Italy). The system provides the 3D coordinates of *retro*-reflective markers placed on the trunk of the subject: 52 on the anterior chest wall surface if the subject laid supine, 47 more markers would be added in the posterior chest wall surface if the subject sat without back support (Cala et al., 1996; Romei et al., 2010). The combination of markers’ coordinates with specific geometrical models and the Gauss’ theorem provides the computation of rib cage (V_{RC}: the volume enclosed from the clavicles to the costal margin), abdominal (V_{AB}: the volume enclosed from the costal margin to the iliac crest) and chest wall (V_{CW}: the sum of the two compartments) volumes.

2.3. sEMG recordings

Respiratory muscles activity was recorded at a sampling rate of 1 KHz using a wireless surface electromyography (BTS FREEEMG; BTS, Milan, Italy) using 8 miniaturized probes with active electrodes. The probes amplify the sEMG signals, digitize them with 16-bit resolution and communicate with the compact receiving unit left on the table and not attached to the subject. The system can be interfaced and synchronized with OEP.

The skin of the subject was prepared by shaving it (when necessary in male subjects), frictioning it with alcohol in order to improve the adherence of the 40 mm diameter foam solid gel surface electrodes positioned on the right side of the subject over the scalene (SCA), parasternal muscles (on the first intercostal space 2 cm apart from the Lewis’ angle, PAR), lower intercostal muscles (8^{th} intercostal space in the mid-axillary line, IC8. This site is indicated to measure also diaphragmatic electrical activity (Verin et al., 2002; Duiverman et al., 2004)), rectus abdominis (on its thoracic insertion 2 cm below the xiphoid process, AXP, and 2 cm lateral to the umbilicus, RAB), external abdominal oblique (between the lower costal margin and the iliac crest 4 cm medially from the nipple line, EOB). Two electrodes were put on the left side over the sternocleidomastoid (SCM). Each couple of electrodes were put at a distance of 4 mm. No reference electrode was positioned as this was a groundless acquisition and the polarization was performed inside each probe.

The sEMG signals were firstly filtered using a band pass digital filter with passband frequency between 10 and 500 Hz and then rectified and the Root Mean Square (RMS) value was computed. RMS during coughs and slow vital capacity was cleaned by subtracting the average RMS value of the spontaneous quiet breathing that preceded the manoeuvres.

A photograph of a subject with the positioning of the sEMG probes and the markers on the anterior chest wall surface is shown on the left panel of Fig. 1.

2.4. Protocol

After a few minutes of spontaneous quiet breathing to rest, subjects were asked to perform a slow vital capacity (SVC) and then to cough as forcefully as possible (4 single voluntary coughs at 40 s intervals) while flow at the mouth, chest wall volumes and sEMG signals were simultaneously recorded. Coughs were executed starting from four different operating volumes randomly ordered: functional residual capacity (FRC), total lung capacity (TLC) and two volumes between TLC and FRC (namely, FRC+ and FRC++) without providing any visual feedback. The protocol was performed before in seated and then in supine position.

The point of start of inspiration, end of inspiration and end of cough have been selected on the chest wall trace to identify the inspiratory and the expiratory cough phases and the thoraco-abdominal contribution was computed for each phase as percentage of V_{CW} variations. (Fig. 1). In this way the inspiratory and expiratory (compressive plus expulsive phase) cough phases were identified (ICP and ECP, respectively) on the ribcage, the abdomen and the chest wall. The root mean square value has been used to quantify the electric signal of each muscle during the two cough phases.

2.5. Statistical analysis

Data distribution of peak cough flow, chest wall volumes and sEMG activity was assessed by Kolmogorov-Smirnov test. If data passed the normality test, they were compared using a one-way repeated measures Analysis of Variance (ANOVA), otherwise the Friedman Repeated Measures ANOVA on Ranks was used. Operating volume and posture were used as independent factors. Post-hoc tests were based on Holm-Sidak andTurkey method in case of parametric and non-parametric test, respectively. Data are expressed as median (25^{th}–75^{th} percentiles). Significance was determined by $p < 0.05$ (SigmaStat 3.5, Systat Software, Inc., San Jose, CA, USA).

3. Results

3.1. Subjects

The complete protocol, in terms of measurements (flow at the

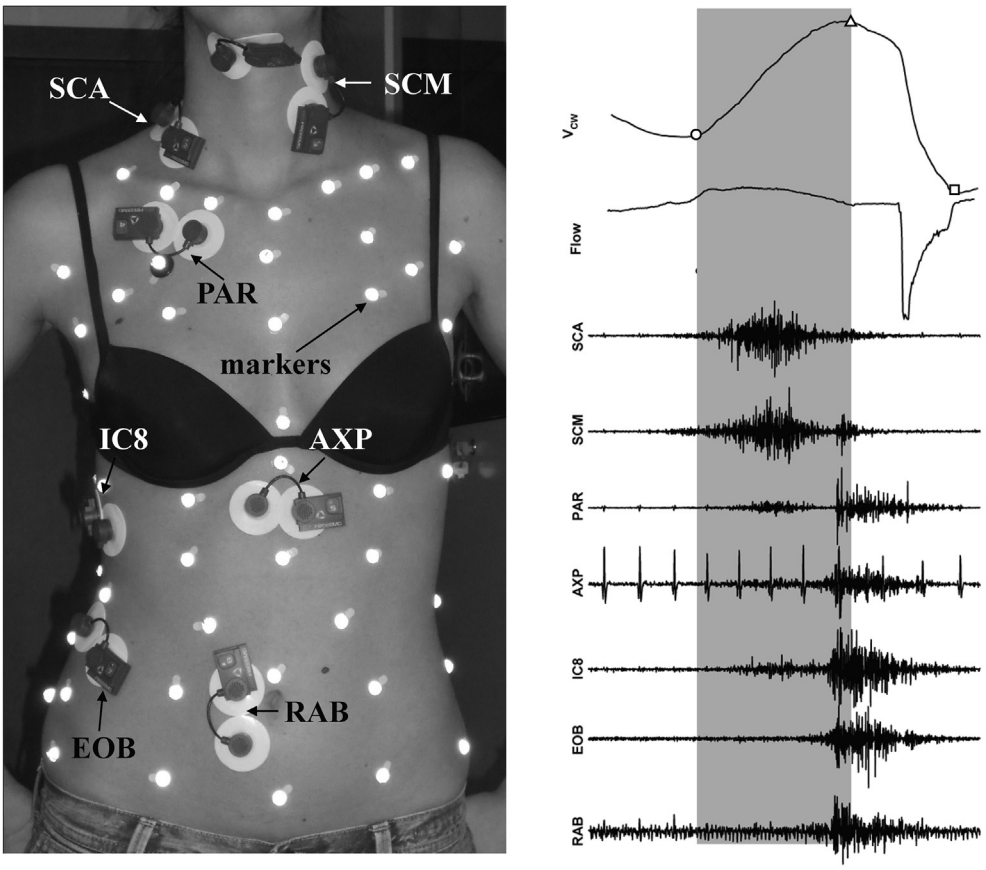

Fig. 1. sEMG electrodes positions and volume, flow and sEMG recordings – Photograph of a subject wearing markers and electrodes for sEMG measurements of the sternocleidomastoid (SCM), the scalene (SCA), the parasternal (PAR), the intercostal (IC8), the external abdominal oblique (EOB) and the rectus abdominis (AXP and RAB) muscles (left panel). Chest wall volume, flow and sEMG tracings of the considered respiratory muscles during a single voluntary cough starting from total lung capacity. The point of start of inspiratory (cirlce), end of inspiratory (triangle) and end of expiratory (square) cough phases are shown on the chest wall trace. The grey area represents the expiratory cough phase that is the sum of the compressive and the expulsive cough phases.

mouth, sEMG and OEP), postures (supine and seated) and operating volumes (FRC, FRC+, FRC++ and TLC), was available in ten (5 females) subjects (age: 23 (22.3–27.3) years; height: 1.74 (1.71–1.77) m; weight: 65 (60–73.8) kg). They were all healthy subjects, self-reporting no important diseases, and with normal forced vital capacity (99.1 (95.6–102) percentage of predicted values computed according to the global lung initiative (Quanjer et al., 2012)).

3.2. Operating volumes and peak cough flow

Despite the lack of a visual feedback, subjects were able to reach four different operating volumes, similar within the two postures, as shown by Table 1 in which the operating volumes are expressed as percentage of the inspiratory capacity of the slow vital capacity that preceded the manouvers. Accordingly, peak cough flows augmented with increasing operating volume and were comparable in seated and supine positions (Table 1).

Tidal volume during the quiet breathing that preceded cough manouver in seated and supine position was, respectively, 25.3 (20.8–32.3) and 20.6 (14.2–23.6) percentage of the inspiratory capacity.

3.3. Chest wall volumes during cough phases and SVC

The points of chest wall start of inspiration, end of inspiration and end of cough are shown in Fig. 2 together with the corresponding points during the slow vital capacity. While end inspiratory cough volume augmented with increasing operating volume approaching TLC as

Table 1
Peak cough flow (PCF) and operating volumes (O.V.) expressed as percentage of the total lung capacity (TLC) during slow vital capacity preceding cough at the four considered O.V.: functional residual capacity (FRC), total lung capacity (TLC) and two volumes between FRC and TLC (namely, FRC+ and FRC++). Data are expressed as median, 25th (P_{25}) and 75th (P_{75}) percentiles.

	PCFI (L/min)				O.V (%IC)			
	Median	P_{25}	P_{75}	*P*-value	Median	P_{25}	P_{75}	p-Value
Seated								
Cough @ FRC	−413	132	256	**vs C, D; °°°	0.9	0.1	7.1	***vs B, C, D
Cough @ FRC+	−382	224	180	**vs D	47.4	42.0	53.3	***vs C, D
Cough @ FRC++	−565	88	199		76.1	48.7	82.0	*** vs D
Cough @ TLC	−652	64	225		94.9	90.5	102.5	
Supine								
Cough @ FRC	−202	−276	−85	***vs C, D	0.7	0.0	2.0	***vs B, C, D
Cough @ FRC+	−149	−479	−73	**vs D	32.6 25.5 38.2	***vs C, D		
Cough @ FRC++	−442	−588	−185		67.4	49.0	75.0	***vs D
Cough @ TLC	−624	−650	−411		103.6	96.8	106.6	

*, **, ***: p < 0.05, 0.01, 0.001; B: operating volume = FRC+; C: operating volume = FRC++; D: operating volume = TLC.

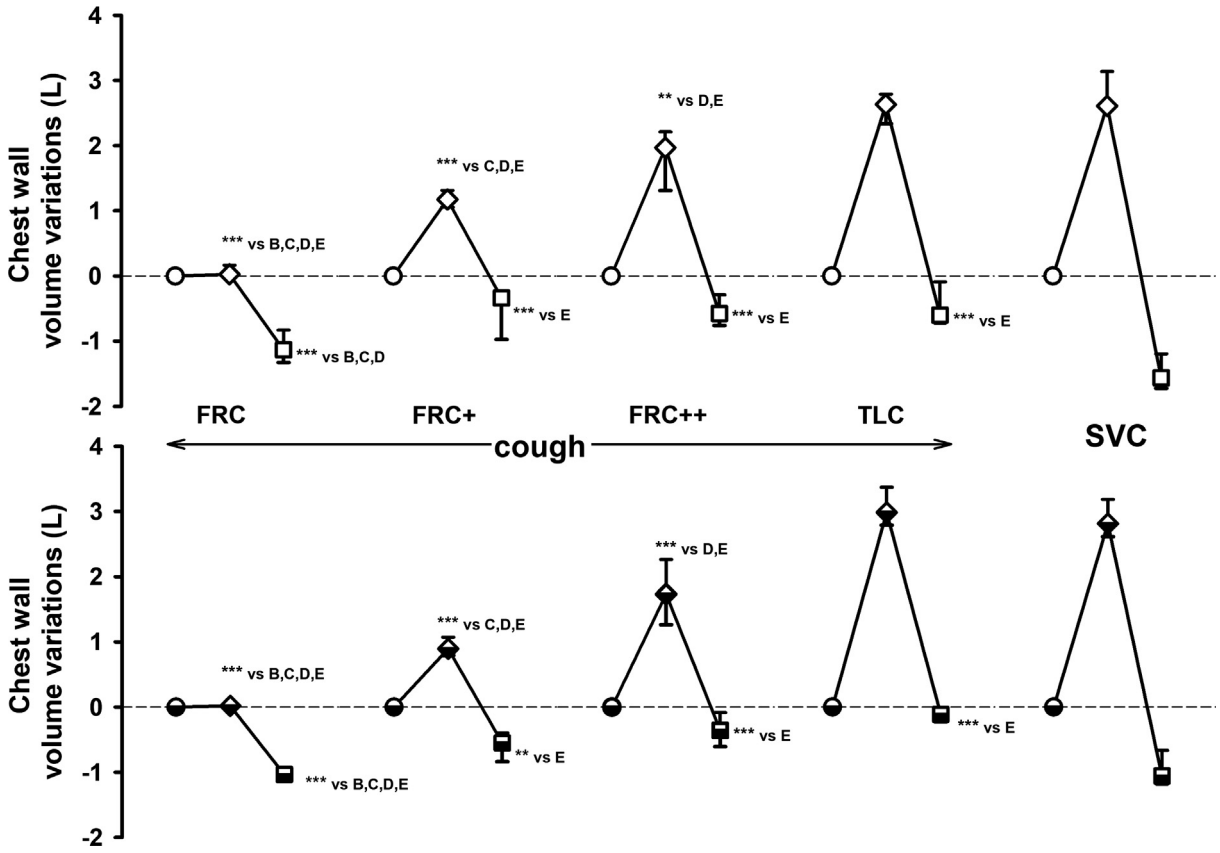

Fig. 2. Chest wall volume during cough – Median (symbols) and interquartile range (whiskers) of chest wall volume at the start of the inspiratory (circles), end of the inspiratory (diamond) and end of the expiratory (square) cough phases at each operating lung volume: functional residual capacity (FRC), total lung capacity (TLC) and two volumes between FRC and TLC (namely, FRC+ and FRC++) in seated (top panel, white symbols) and supine (bottom panel, semi-filled bottom symbols) positions. The corresponding points during slow vital capacity (SVC) are also reported. The one-way repeated measures Analysis of Variance (ANOVA) was used when the whiskers were symmetric with respect to the median, indicating normal data distribution. The Friedman Repeated Measures ANOVA on Ranks was used otherwise. **,***: $p < 0.01, 0.001$; B: operating volume = FRC+; C: operating volume = FRC++; D: operating volume = TLC; E: SVC manouver.

expected, end expiratory cough volume significantly reduced coughing from TLC to FRC. In the latter the volume at the end of cough almost reached the end of SVC, *i.e.*: the residual chest wall volume.

The thoraco-abdominal percentage contribution during ICP and ECP revealed that both phases turned to be predominantly thoracic as shown in Fig. 3.

Interestingly, operating volumes had a slight effect on the thoracic predominance during cough only in ECP. On the other hand, the change of posture did influence the thoracic contribution during ICP and during the quiet breathing that precedes cough. In fact, it significantly ($p < 0.001$) reduced passing from seated (70 (62.8–75.4)%) to supine (39.0 (29.6–52.6)%) position.

3.4. RMS of respiratory muscles during cough and SVC

The RMS of the muscles of the neck, of the thorax and of the abdomen during cough and slow vital capacity are shown in Figs. 4–6, respectively.

The activity of scalene and sternocleidomastoid muscles resulted significantly higher only during cough from TLC and during SVC manoeuvres, particularly in the ICP (Fig. 4).

During ECP the activity of the respiratory muscles measured at 8th intercostal space was similar within the different OV and significantly higher in seated position, being reduced at lower OV, while changes were found during the inspiratory effort. No differences were found in the activity of the parasternal with OV, while the ECP in seated position was characterized by higher muscles activity (Fig. 5).

The external abdominal oblique muscle was found to have higher ECP activity when cough started at TLC, regardless OV. On the other hand, significantly higher muscles activity of the rectus abdominis was found in the expiratory phase during cough at lower operating volumes. (Fig. 6).

4. Discussion

This study is the first to measure muscles activation and action during voluntary cough by combining surface electromyography and chest wall kinematics and by investigating the effect of operating volume and posture in healthy human subjects. We have shown that posture did not influence volume variations and muscles activation and that thoraco-abdominal compartments contributed constantly to volume changes during inspiratory and expiratory cough phases, that turns to be mostly thoracic, regardless of the operating volume and posture.

It is well known that posture influences thoraco-abdominal kinematics during quiet breathing, being predominantly abdominal in supine position and thoracic in seated (Romei et al., 2010). This was confirmed by the data of tidal volume that preceded cough: rib cage contribution significantly decreased when adopting the supine position. For this reason, we would have expected two different patterns (one for each posture) also during coughing. Although the supine position introduces mechanical changes in the chest wall (*i.e.* cranially displacement of the diaphragm, lung volume reduction, stretching of the expiratory muscles and lowering of the abdominal compliance), surprisingly, peak cough flow, chest wall volumes and muscular activation resulted similar with posture both in ICP and ECP. In addition,

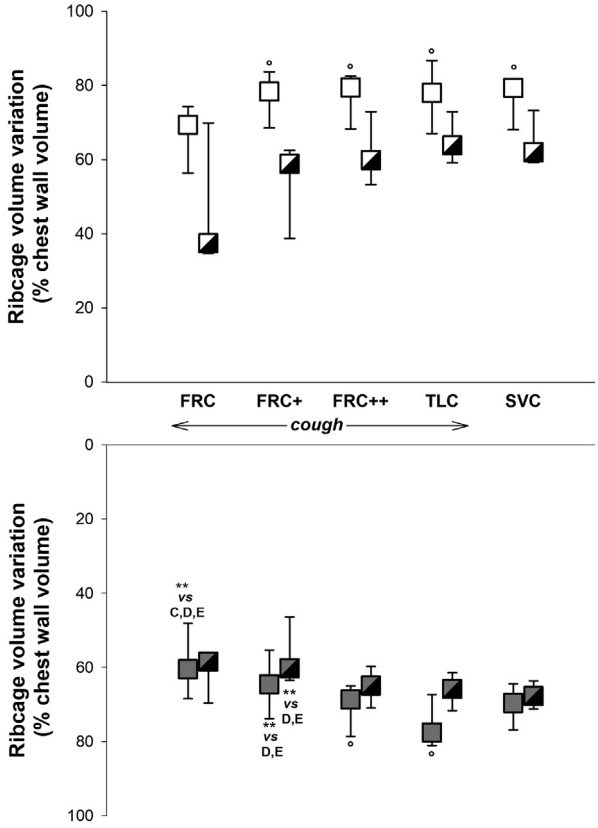

Fig. 3. Ribcage contribution –Median (symbols) and interquartile range (whiskers) of the percentage contribution of the rib cage during the inspiratory (top panel, white symbols) and expiratory (bottom panel, grey symbols) cough phases at each operating lung volume: functional residual capacity (FRC), total lung capacity (TLC), two volumes between FRC and TLC (namely, FRC+ and FRC++) and during slow vital capacity (SVC) in seated (open symbols) and supine (semi-filled bottom symbols) positions. The one-way repeated measures Analysis of Variance (ANOVA) was used when the whiskers were symmetric with respect to the median, indicating normal data distribution. The Friedman Repeated Measures ANOVA on Ranks was used otherwise. **: $p < 0.01$; B: operating volume = FRC+; C: operating volume = FRC++; D: operating volume = TLC; E: SVC manouver. °: $p < 0.05$ vs supine position.

rib cage and abdomen invariantly contributed to both the considered cough phases, with the former being the leading compartment, irrespective of the operating volume and, above all, the posture.

These results suggested that under normal physiological conditions, in order to provide efficient expulsive flow, voluntary cough was associated with a specific co-ordinate muscle activity resulting in a constant thoracic pattern no matter the quantity of inhaled air and/or the posture were. The invariant relative contribution of each compartment during cough at different operating volumes was also found by Smith et al. who investigated cough, but only in seated position (Smith et al., 2012).

It can be speculated that the central control mechanism involved in voluntary cough (Tomori and Widdicombe, 1969; Fontana, 2003; Simonyan et al., 2007; Mazzone et al., 2011) is able to bypass the mechanical changes consequent to postural change but also to increased lung volume (*i.e.* maximal passive elastic recoil during expiration).

Even though cough is an expiratory maneuver and the principal expiratory muscles lie in the ventro-lateral aspect of the abdominal wall, the rib cage resulted the leading compartment during cough, indicating not only an important contribution of the intercostal muscles but also that abdominal muscles mainly act on lowering the rib cage on which they are inserted (Roussos, 1995).

In fact, the sEMG measured at the eighth intercostal space and the one coming from the external oblique muscle during ECP resulted similar within posture and operating volumes but significantly higher than during slow vital capacity, indicating that these muscles seem to be the main responsible for providing efficient voluntary cough in humans.

Coughing at low operating volumes made the activity of the rectus abdominis, measured at its umbilical level, to be higher compared to the contraction needed to reach the residual volume during the slow vital capacity. On the other hand, neck muscles increased their activity working at TLC in both maneuvers, cough and slow vital capacity. This happened both during inspiration, to help lifting the ribs to reach the highest operating volume, and expiration, maybe to stabilize the head and the neck during the expiratory effort.

The high variability of the muscular activity measured at the second intercostal space in seated position, during ECP, might be attributed to the gender of the subjects. In fact, despite the lack of statistical significance mainly due to the low number of volunteers, men seemed to be characterized by higher activity than women. This could be explained by the pectoralis major muscle being more superficial and maybe more fitted in our male subjects. This hypothesis is reasonable since the important active role of this muscle during voluntary cough in healthy and tetraplegic subjects has already been pointed out by other researchers (Estenne and De Troyer, 1990; Fujiwara et al., 1999; Lasserson et al., 2006).

One of these authors (Lasserson et al., 2006) showed that voluntary cough in humans is characterized by a different motor activation mechanism compared to reflex cough. It would be interesting to verify if voluntary and reflex cough differ not only in muscles activity but also in their action on the chest wall, in particularly, to study whether the thoracic pattern is maintained also during reflex and evoked cough. Interestingly, the pattern that we found was maintained also coughing at FRC, with a reduced amount of air in the lung to be compressed and exhaled that made the expiratory muscles length to be less optimal. The only difference found, compared to higher operating volumes, was a stronger recruitment of the expiratory reserve volume: the volume reached at the end of ECP, in fact, approached the residual volume reached at the end of slow vital capacity. Coughing at FRC, with its lack of inspiratory phase followed by the forced expiratory effort against a closed glottis, resembled the expiratory reflex, the function of which is to prevent aspiration of foreign material into the airways (Tatar et al., 2008). The expiratory reflex occurs when the stimuli comes from the vocal folds or upper tracheal segments (Fontana and Lavorini, 2006) and therefore it should play an important role in case of general anaesthesia (Drummond et al., 2011) or chocking, particularly in very severe patients. In particular, we have recently shown that the resistance in the tracheal tube seemed to trigger an active expiration/coughing respiratory pattern that occurred to restore breathing during emergence (Kostic et al., 2018).

It would be interesting to check whether the thoracic pattern was maintained also in patients with ribcage alteration (*i.e.*: severe scoliosis, pectus exavatum, osteogenesis imperfecta or post thoracic surgery) and muscular problems (*i.e*: neuromuscular diseases, paralysis or hemiplegia) and/or if they adopted a compensatory strategy to try to get efficient cough. We have studied chest wall volumes in supine position during voluntary cough in severe Duchenne muscular dystrophy (DMD) patients whose peak cough flow was lower than 160 L/min making their cough inefficient. The thoracic pattern was maintained in these patients because rib cage muscles were weakened to a lesser extent and they had to lead ventilation and cough manoeuvre in disadvantage condition, due to the supine position. Intercostal muscles passed form being accessory to control respiration in order to compensate the impaired diaphragm but their action resulted in poor chest wall expansion. The role of the diaphragm, therefore, was crucial also during cough because it contributed to the inspired volume preceding coughing, the operating volume, that determines not only the volume of

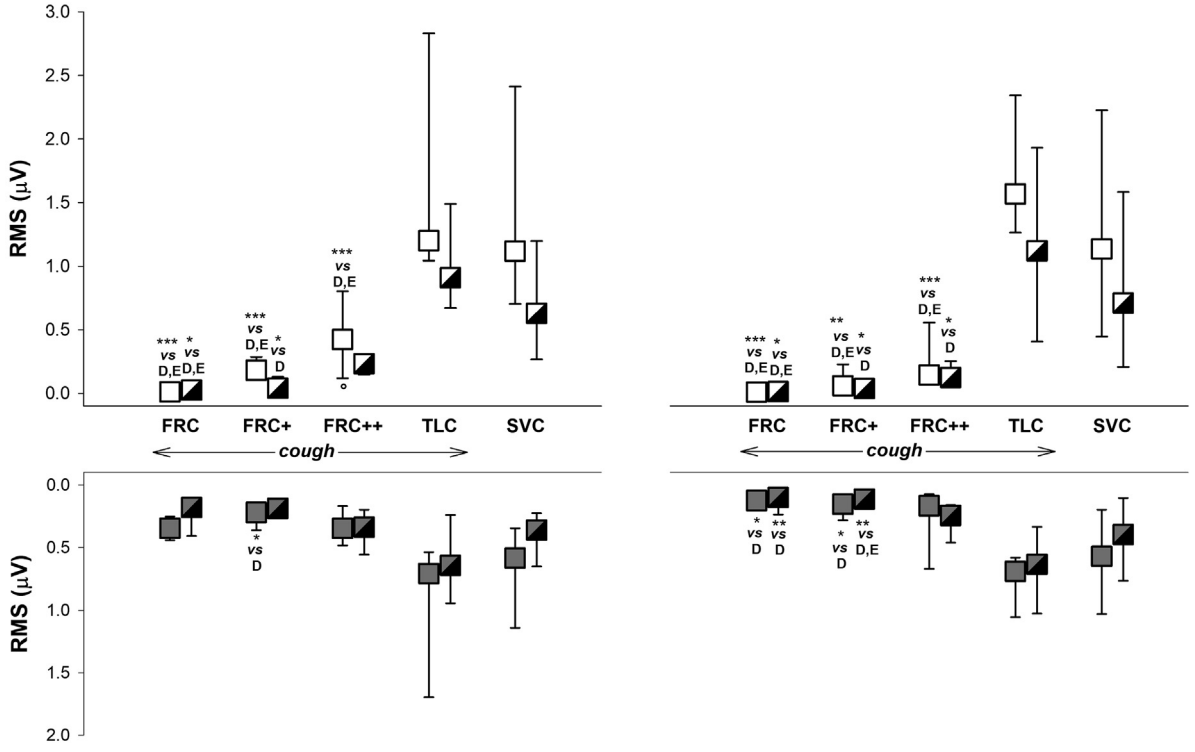

Fig. 4. RMS of accessory neck muscles –Median (symbols) and interquartile range (whiskers) of root mean square value of the sEMG signals for SCA (left panels) and SCM (right panels) muscles in seated (open symbols) and supine (semi-filled bottom symbols) positions during the inspiratory (top panels, white symbols) and the expiratory (bottom panels, grey symbols) cough phases at each operating volumes: functional residual capacity (FRC), total lung capacity (TLC), two volumes between FRC and TLC (namely, FRC+ and FRC++) and during slow vital capacity (SVC). The one-way repeated measures Analysis of Variance (ANOVA) was used when the whiskers were symmetric with respect to the median, indicating normal data distribution. The Friedman Repeated Measures ANOVA on Ranks was used otherwise. *, **, ***: $p < 0.05, 0.01, 0.001$; D: operating volume = TLC; E: SVC manouver.

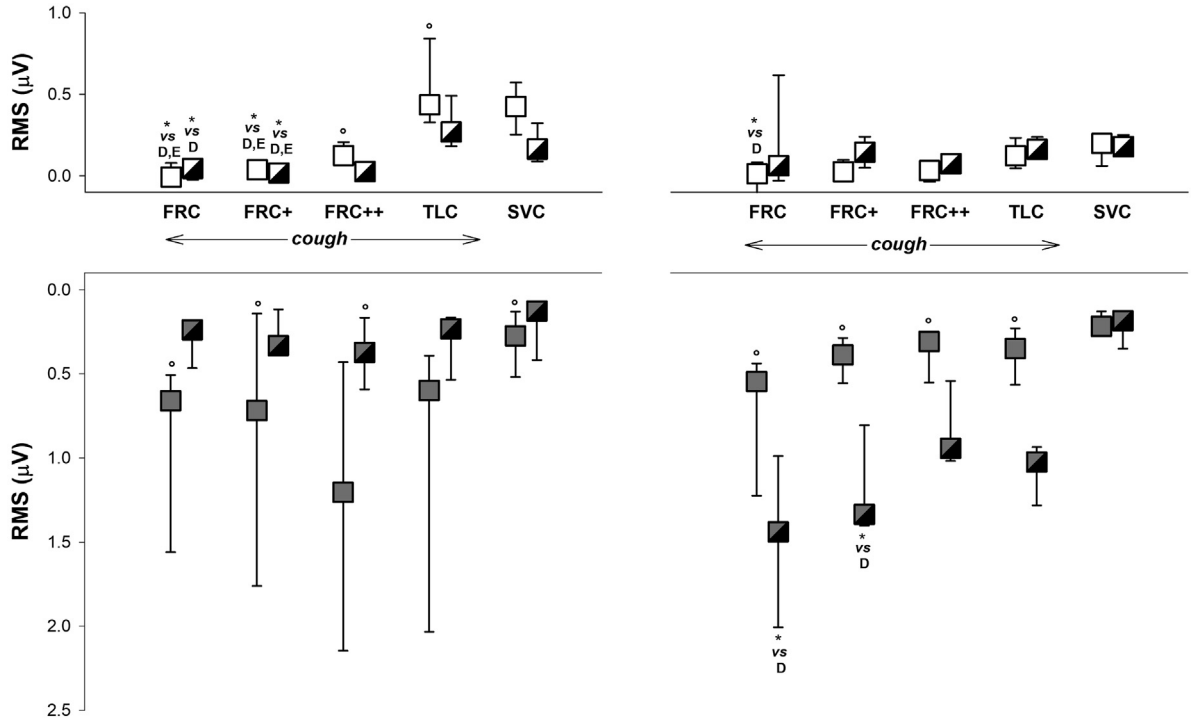

Fig. 5. RMS of thoracic muscles –Median (symbols) and interquartile range (whiskers) of root mean square value of the sEMG signals for PAR (left panels) and IC8 (right panels) muscles in seated (open symbols) and supine (semi-filled bottom symbols) positions during the inspiratory (top panels, white symbols) and the expiratory (bottom panels, grey symbols) cough phases at each operating volumes: functional residual capacity (FRC), total lung capacity (TLC), two volumes between FRC and TLC (namely, FRC+ and FRC++) and during slow vital capacity (SVC). The one-way repeated measures Analysis of Variance (ANOVA) was used when the whiskers were symmetric with respect to the median, indicating normal data distribution. The Friedman Repeated Measures ANOVA on Ranks was used otherwise. *: $p < 0.05$; °: $p < 0.05$ vs supine position; D: operating volume = TLC; E = SVC manouver.

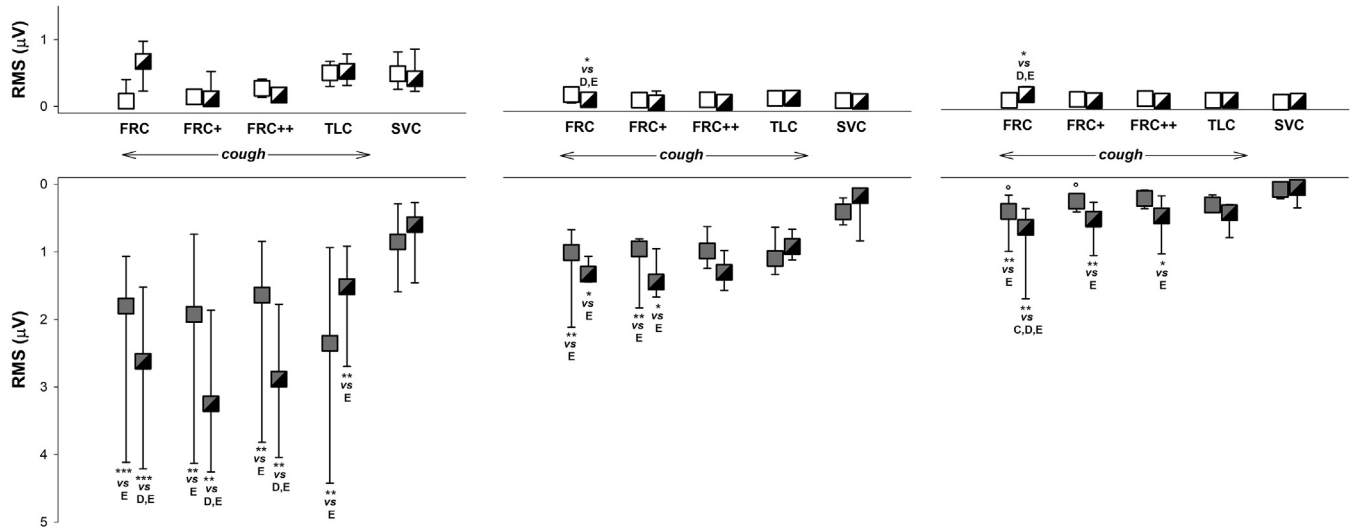

Fig. 6. RMS of abdominal muscles –Median (symbols) and interquartile range (whiskers) of root mean square value of the sEMG signals for AXP (left panels), EOB (middle panels) and RAB (right panels) muscles in seated (open symbols) and supine (semi-filled bottom symbols) positions during the inspiratory (top panels, white symbols) and the expiratory (bottom panels, grey symbols) cough phases at each operating volumes: functional residual capacity (FRC), total lung capacity (TLC), two volumes between FRC and TLC (namely, FRC+ and FRC++) and during slow vital capacity (SVC). The one-way repeated measures Analysis of Variance (ANOVA) was used when the whiskers were symmetric with respect to the median, indicating normal data distribution. The Friedman Repeated Measures ANOVA on Ranks was used otherwise. **, **: $p < 0.05, 0.01$; C: operating volume = FRC++; D: operating volume = TLC; E = SVC manouver.

air that can be expelled but also the length-tension relationship of the expiratory muscles, *i.e.* their ability of producing force (LoMauro et al., 2014a). We have also studied ICP during voluntary cough in adult patients with spinal muscular atrophy (SMA) type II, the most severe non-lethal form, whose diaphragm is relatively preserved while the rib cage muscles are compromised. These patients were characterized by an altered kinematic compensatory pattern, being predominantly abdominal, during the inspiratory and the expiratory cough phase (LoMauro et al., 2014b). Both type II SMA and severe DMD patients used assisted cough device because their cough was inefficient underling how performing efficient cough is a combination of adequate operating volume, thoracic kinematics and respiratory muscles activation. The role of the diaphragm during cough was studied mainly on anaesthetized animals. Because the sEMG measured at the eighth intercostal space is suggested to be the best indicator of the activity of the diaphragm via transcutaneous electromyography, our IC8 results during the inspiratory phase could add more information on the diaphragmatic activity during cough. The diaphragm seemed to contribute constantly to reach all the four OV that preceded cough but also during the inspiratory maneuver that preceded slow vital capacity in both positions (Lasserson et al., 2006; Vovk et al., 2007; Hegland et al., 2012). This results further confirmed the important role of the so called accessory muscles in maneuver other than quiet breathing, with the diaphragm being “only” responsible for resting breathing.

4.1. Critique of the methods

We considered the root mean square, *i.e.* the integration of the total electrical activity during contraction in both the considered cough phases, instead of the “moving average” of the signal which takes in consideration also the duration of the muscular effort. Moreover, we have normalized the RMS values to the quiet breathing preceding cough and not to peak amplitude of maximal muscle activation. Because, a part from the diaphragm, all the other considered muscles are not “purely” respiratory but they all have other function, mainly postural, the choice of the maximal reference values is crucial. In fact, it has been shown that abdominal and intercostal accessory muscles can be driven to a greater degree during voluntary postural efforts and not during respiratory ones (Gandevia et al., 1990). We are also aware of the limitation of the transcutaneous EMG measurements which do not reflect the activation of a single muscle, but rather, that of different muscular layers with an important cross-talk of the adjacent muscles. For the same reason, the study of deepen muscles, like the diaphragm, can be very poor with sEMG. We have considered ECP as a whole, and not split into the compressive phase and expulsive phase, because the quality of the flow signal during the compressive phase was not low and we have preferred to use the volume signal that allowed only to distinguish between the inspiratory and expiratory phases.

We have not considered coughs below FRC that is associated with increased sEMG compared with higher OV. This would be helpful for future studies of peals cough, that typically occurs in chronic respiratory disease and/or to clear the airways of mucus and irritants. Operating volumes progressively reduce with the end of long peals of coughing that is likely to occur below FRC (McGuinness et al., 2018). Finally, another important factor that we have not considered but that has a strong influence is cough effort. McGuinness and colleagues, in fact, showed that the effect of the effort is higher than OV not only on sEMG, but also on thoraco-abdominal pressures and sound produced (McGuinness et al., 2018).

On the other hand, our results were in line with already published data. In particular, they confirmed the important role of the external abdominal oblique muscles during the expulsive phase of cough in humans (Cox et al., 1984; De Troyer et al., 1990; Floyd and Silver, 1950; Fontana et al., 1997; Strohl et al., 1981). The rectus abdominis is known to contribute with a smaller activation (Floyd and Silver, 1950; Strohl et al., 1981) and this was confirmed by our data only at higher operating volumes. The rectus abdominis vigorously activated when coughing occurred at low lung volume, therefore supporting the action of the external oblique muscle to recruit the expiratory reserve volume in order to guarantee an efficient expulsive phase. In supine position the rectus abdominis tended to increase its electrical activity measured at its xiphoydeal insertion maybe thanks to the beneficial stretching effect of the posture. The general increased sEMG at highest operating volume, that we found, was confirmed also independently on cough effort (McGuinness et al., 2018). Finally, the invariant contribution of the diaphragm at different operating volumes was already found by measuring the trans- diaphragmatic pressure (Smith et al., 2012) and therefore in a more invasive method.

In conclusion, we combined transcutaneous sEMG measurements with the kinematics of the chest wall to study the inspiratory and the expiratory phase of voluntary cough in healthy subjects. The synergetic antagonistic activity of respiratory muscles leads to a specific pattern, in which the rib cage is the leading compartment indicating that the abdominal muscles mainly act to lower the inferior ribs due to their insertions on the rib cage. This thoracic pattern is not affected by either operating volume or posture despite the mechanical changes they introduced and this suggest a task-specific somatosensory activation during voluntary cough.

Declaration of Competing Interest

The authors declared that there is no conflict of interest.

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