Received: 2 September 2023 | Accepted: 31 January 2024

DOI: 10.1002/ppul.26915

ORIGINAL ARTICLE

PEDIATRIC PULMONOLOGY WILEY

Short-term effects of positive expiratory pressure mask on ventilation inhomogeneity in children with cystic fibrosis: A randomized, sham-controlled crossover study

Simone Gambazza PhD[1] | Alessandra Mariani MSc[2] | Riccardo Guarise PT[3] | Beatrice Ferrari PT[4] | Federica Carta PT[2] | Anna Brivio MSc[1] | Sofia Bizzarri PT[4] | Chiara Castellani PT[4] | Carla Colombo MD[2,5] | Dario Laquintana MSc[1]

[1]Healthcare Professions Department, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

[2]Cystic Fibrosis Centre, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

[3]Cystic Fibrosis Centre, University Hospital of Verona, Verona, Italy

[4]Rehabilitation Unit, Meyer Children's Hospital IRCCS, Florence, Italy

[5]Department of Pathophysiology and Transplantation, Università degli Studi di Milano, Milan, Italy

Correspondence

Simone Gambazza, PhD, Healthcare Professions Department, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Via Francesco Sforza 35, Milan 20122, Italy.
Email: simone.gambazza@policlinico.mi.it

Funding information

This study was partially funded by the Italian Ministry of Health—Current research IRCCS

Abstract

Background: Can physiotherapy with a positive expiratory pressure (PEP) mask improve peripheral ventilation inhomogeneity, a typical feature of children with cystic fibrosis (cwCF)? To answer this question, we used the nitrogen multiple-breath washout (N_2MBW) test to measure diffusion-convection-dependent inhomogeneity arising within the intracinar compartment (S_{acin}*VT).

Methods: For this randomized, sham-controlled crossover trial, two N_2MBW tests were performed near the hospital discharge date: one before and the other after PEP mask therapy (1 min of breathing through a flow-dependent PEP device attached to a face mask, followed by three huffs and one cough repeated 10 times) by either a standard (10–15 cmH_20) or a sham (<5 cmH_20) procedure on two consecutive mornings. Deception entailed misinforming the subjects about the nature of the study; also the N_2MBW operators were blinded to treatment allocation. Study outcomes were assessed with mixed-effect models.

Results: The study sample was 19 cwCF (ten girls), aged 11.4 (2.7) years. The adjusted S_{acin}*VT mean difference between the standard and the sham procedure was −0.015 (90% confidence interval [CI]: −∞ to 0.025) L^{-1}. There was no statistically significant difference in S_{cond}*VT and lung clearance index between the two procedures: −0.005 (95% CI: −0.019 to 0.01) L^{-1} and 0.49 (95% CI: −0.05 to 1.03) turnovers, respectively.

Conclusion: Our findings do not support evidence for an immediate effect of PEP mask physiotherapy on S_{acin}*VT with pressure range 10–15 cmH_20. Measurement with the N_2MBW and the crossover design were found to be time-consuming and unsuitable for a short-term study of airway clearance techniques.

KEYWORDS

airway clearance technique, cystic fibrosis, multiple-breath washout, physiotherapy

Alessandra Mariani and Riccardo Guarise contributed equally to this study.

1 | INTRODUCTION

Physiotherapy in cystic fibrosis (CF) comprises airway clearance techniques (ACTs), exercise testing and training, inhalation therapy, and life-long education for children with cystic fibrosis (cwCF) and their caregivers.[1,2] Some ACTs are based on the use of positive expiratory pressure (PEP). Heterogeneity in study design and endpoints has limited the internal and external validity of evidence about PEP breathing effect in CF. A systematic literature review reported that short- and long-term studies found no significant difference between PEP and other ACTs for improving pulmonary function, exercise capacity, or quality of life.[3] Longer-term studies comparing PEP with other techniques showed equivocal or conflicting results for forced expiratory volume in the first second (FEV_1).[3,4] A significant reduction in respiratory exacerbations requiring antibiotics was found in patients who received PEP compared with those who received high-frequency chest wall oscillation over a 1-year period.[5] These endpoints are relatively insensitive to evaluating airway clearance with PEP mask therapy in cwCF, however.

The rationale behind regular application of PEP breathing is to temporarily increase functional residual capacity (FRC) by breathing through a closed system,[6] and to increase interdependence between the alveoli, thus facilitating collateral ventilation[7] and recruiting previously obstructed airways.[8] As airway pressure increases during expiration, premature collapse is prevented and gas trapping in the lungs reduced,[9] resulting in modified distribution of ventilation—theoretically. The diffusion-convection (S_{acin}*VT), the convection (S_{cond}*VT) dependent indices, and the lung clearance index (LCI) measured with the multiple-breath washout (MBW) test reflect ventilation inhomogeneity arising from the intracinar compartment, within the airways proximal to the terminal bronchioles and global ventilation efficiency, respectively. Among currently available lung functional tests, the MBW test allows for discriminating between preschool cwCF and healthy controls by virtue of the regionally heterogeneous nature of early airway obstruction.[10,11] In the past 10 years, LCI has been increasingly used as a short-term endpoint to determine the efficacy of physiotherapy in cwCF[12–17] but has produced conflicting results. Under ideal and purely theoretical conditions, all lung units empty simultaneously during expiration, whereas if gas mixing deficits are present, the nitrogen (N_2) from the areas of patchy disease is expired late and creates a certain phase III slope because healthier respiratory units are washed out first. These variations in the rate and amount of emptying in CF occur in multiple lung branches due to uneven narrowing of parallel airways caused by disease processes or differences in airway branch volumes. Hence the reason to use ventilation inhomogeneity as a more sensitive endpoint to evaluate the short-term effects of PEP mask therapy in CF. Under such conditions, we expected a change in S_{acin}*VT due to PEP breathing that would be theoretically and physiologically justifiable.

To our best knowledge, no trial so far has investigated the immediate effect of ACTs on MBW-derived S_{acin}*VT, which was found a sensitive endpoint to CF transmembrane conductance regulator (CFTR) modifying treatment.[18] For this study involving a sham comparator and deception of subjects and their caregivers about the study purpose, we wanted to determine the extent to which PEP mask therapy may decrease peripheral ventilation inhomogeneity in cwCF. To do this, we applied PEP breathing (pressure range 10–15 cmH_20) and measured the changes in the diffusion-convection dependent index S_{acin}*VT.

2 | METHODS

2.1 | Study design and population

This multicenter, randomized sham-controlled crossover trial (ClinicalTrials.gov identifier: NCT03760120) was conducted at two study sites, the Cystic Fibrosis Center, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, and the Meyer Children's Hospital IRCCS, Florence. The study is presented in conformity with CONSORT guidelines.[19]

The subjects underwent two tests in a two-period, two-sequence crossover design (Figure 1). On the 1 day, they were randomized to receive the first intervention, followed by a second intervention after a 1-day washout period. All tests were performed in the morning. They were randomized 1:1 to the orders of the two study arms in a standard-sham/sham-standard sequence according to a computer-generated allocation schedule, which was generated by one member of the CF staff not involved in the study.

Children and adolescents diagnosed with CF based on a positive sweat test (chloride > 60 mEq/L) and/or the presence of two disease-causing mutations were consecutively recruited. The inclusion criteria were: use of a PEP mask as their routine ACT, age between 5 and 18 years, body weight ≥ 15 kg, predicted FEV_1 ≥ 40%, ability to perform the N_2MBW test, and spirometry. Exclusion criteria were: signs and/or symptoms of respiratory exacerbation[20] within the previous 2 weeks, awaiting lung transplantation, and receiving noninvasive mechanical ventilation or oxygen therapy.

The cwCF were screened near their hospital discharge to determine eligibility and to assess ongoing inhaled therapies, which would be discontinued for the duration of the study. Before each procedure, the subjects were instructed not to take short-acting beta-agonists or anticholinergics for 6 h and long-acting beta-agonists or anticholinergics for 12 h and not to perform their usual morning airway clearance with a PEP device.

2.2 | Lung function testing

Spirometry was performed during the screening visit according to American Thoracic Society (ATS)/European Respiratory Society (ERS) guidelines.[21] FEV_1 was converted into a *Z* score.[22] An open-circuit MBW hard- and software package with nitrogen as tracer gas (N_2MBW) was used and data from earlier software versions were re-run (Exhalyzer® D and Spiroware 3.3.1 Ecomedics AG); calibration and measurement were performed according to the manufacturer's instructions.[23–26] The results from three reproducible runs were recorded, based on currently available

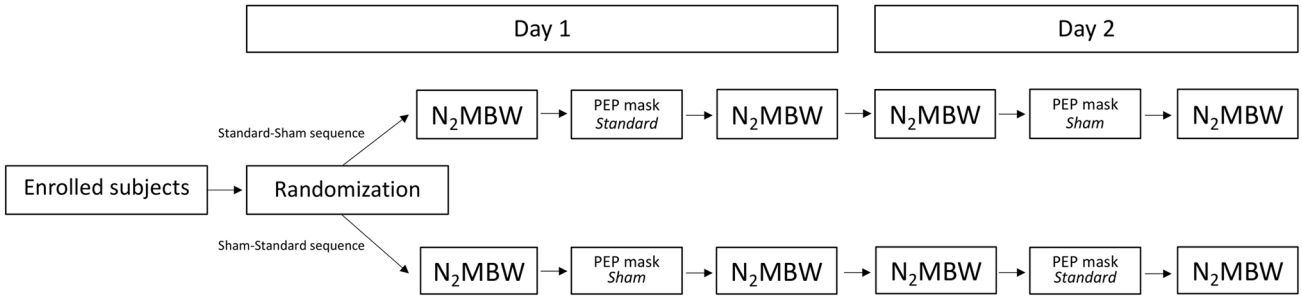

FIGURE 1 Two period, two-sequence crossover design. N_2MBW denotes nitrogen multiple-breath washout. PEP, positive expiratory pressure.

quality criteria.[26] The N_2MBW test following the standard or the sham intervention was performed after twice the washout time recorded for each subject at the baseline N_2MBW test. This was done to be sure that the nitrogen levels would return to baseline end-tidal values.[27,28] The tests were performed in an environment providing adequate distraction for young children.[25] The N_2MBW operators were blinded to treatment allocation.

2.3 | PEP breathing intervention

The standard and the sham intervention entailed the application of a PEP mask under the supervision of a physiotherapist specialized in CF care. The PEP mask is a flow-resistor device in which the outflow resistance at a constant expiratory flow is inversely correlated to the diameter of the outflow resistor. The PEP mask consisted of a silicon face mask to which resistors (inner diameter: 1.5–5.0 mm) can be connected. The resistor was connected to a unidirectional valve (PEP/Rmt™). The PEP mask was applied with the subject seated on an adjustable chair at a table, with knees and hips bent to 90°, feet firmly placed on the floor, elbows resting on the table, and the subjects holding the mask with their hands. The subjects had used a PEP mask since diagnosis of CF in their first months of life. As per clinical practice, PEP mask therapy is applied twice a day, therefore, no training was necessary for this study sample.

The standard and the sham intervention entailed active but not forced breathing through the mask for 1 min followed by a brief pause (30 s–1 min) during which the subject huffed through a mouthpiece three times. The huffing maneuver consists of forced exhalation from total lung capacity to residual functional capacity plus one cough. The intervention was repeated 10 times, to achieve 10 min of PEP breathing.[6,29]

The airflow resistor in the standard intervention generated a pressure range 10–15 cmH_20, while a large resistor that generated <5 cmH_20 (not considered effective) was applied in the sham intervention.[6,30] A manometer inserted between the valve and the outlet ensured that all subjects generated the right amount of pressure and that the working pressure was visible only to the physiotherapist.

2.4 | Outcomes

The primary outcome was S_{acin}*VT at the end of each intervention. The secondary outcome measures were S_{cond}*VT, LCI, and sputum wet weight. Sputum produced spontaneously during and after each intervention (standard and sham) was collected in a sterile, preweighed container (balance precision 0.1 g). Oxygen saturation (SpO_2) was monitored throughout as a safety measure. The recruitment pace, defined as the mean number of subjects recruited over the study period (years), was the measure of feasibility.

2.5 | Sample size

Previous studies at the Milan study site (unpublished data) using the N_2MBW test showed a mean standard deviation (SD) of difference of 0.073 L^{-1} in the S_{acin}*VT between pre- and post-PEP mask therapy. A total of 18 subjects were planned to enter this crossover study. There was a 90 percent probability that the study would detect a difference between treatments at a one-sided 0.05 significance level if the true difference between treatments is 0.053 L^{-1}, which defines a 20% improvement.

2.6 | Statistical analysis

All variables are described using mean and ±1 SD or count and percentage. A mixed linear model was based on the fixed effects of intervention, intervention sequence, study center, and baseline value, and a random effect for participants to account for the crossover design.[31] Point estimates from the fitted model are reported as the mean difference between interventions with a two-sided 95% confidence Interval (CI), except for the primary outcome, the precision of which is reported as a one-sided 90% CI. The sensitivity analysis compared the interaction between intervention and FEV_1 based on the lower limit of normal (LLN) to test whether differences between the interventions could be due to the degree of airflow obstruction. The paired Wilcoxon test was used to compare the difference in sputum wet weight between the interventions. All analyses were performed with interventions labeled as A and B,

recoded as sham or standard after finalization. Statistical significance was set at a $p < .05$. All statistical tests were performed using the open-source software R Core Team, version 4.0.3.[32] with packages *lmer* and *emmeans* added.

2.7 | Ethical considerations

The deception involved giving an inaccurate account of the study purpose and a false description of the sham intervention: the subjects were told that the study aim was to determine a more effective level of PEP pressure in changing ventilation inhomogeneity. The consent form explicitly stated that certain aspects of the study would be intentionally misdescribed and that on completion of the study, the investigator would debrief the subjects and their caregivers. The study was approved by the local ethics committee of the Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico (2291/2016), and written informed consent was obtained from the subjects' parents or legal guardians. All research was conducted in accordance with relevant guidelines[33] and the tenets of the Declaration of Helsinki.

3 | RESULTS

The study sample was 19 cwCF enrolled between November 2016 and May 2021 (Figure 2). The study was suspended from March 2020 to February 2021 because of COVID-19 restrictions. Baseline FEV_1 was above the −1.64 *Z* score in 10 cwCF (52.6%) and the LCI was <7.1 in one (5.3%). FEV_1 was lower in the standard-sham sequence compared with the sham-standard, whereas S_{acin}*VT was higher. Table 1 presents study sample characteristics stratified by treatment sequence.

Within- and between-sequence comparison revealed no neat pattern of change in S_{acin}*VT after either standard or sham PEP mask therapy (Figure 3). The mean change in S_{acin}*VT was 0.008 (95% CI: −0.038 to 0.053) L^{-1} after the standard and 0.022 (95% CI: −0.038 to 0.082) L^{-1} after the sham intervention; the resulting adjusted effect of standard PEP mask therapy was −0.015 (90% CI: −∞ to 0.025) L^{-1}. Sensitivity analysis disclosed no difference in FEV_1 LLN between the standard and the sham intervention: S_{acin}*VT was −0.013 (95% CI: −0.110 to 0.085) L^{-1} lower after the standard compared with the sham intervention when FEV_1 was <LLN and −0.02 (95% CI: −0.116 to 0.076) L^{-1} lower when FEV_1 was ≥LLN.

Analysis of secondary outcomes showed no difference between the two interventions (Table 2). Wet sputum weight after the standard procedure was between 0.0 and 10.7 g and between 0.0 and 7.6 g ($p = .295$) after the sham procedure. The change from baseline SpO_2 after the standard and the sham procedure was 0.5 (95% CI: −0.1 to 1.1)% and 0.5 (95% CI: −0.2 to 1.2)%, respectively. There was no difference in FRC between the procedures: 0.04 (95% CI: −0.12 to 0.21) L. No adverse events were recorded during the study. The pace of recruitment was 2.8 subjects per year (over 5 years).

4 | DISCUSSION

This randomized crossover trial found insufficient evidence to support a 20% improvement in ventilation inhomogeneity arising from the intracinar compartment after PEP mask therapy delivered at a pressure range of 10–15 cmH_20 as compared with therapy delivered at a pressure range <5 cmH_20. There were no differences in the derivatives of the N_2MBW test between the two procedures in this sample.

Routine use of a PEP mask therapy for airway clearance in CF rests on the technique's ability to increase FRC and to improve airflow passage in obstructed small airways through collateral ventilation, thus preventing premature airway collapse and increasing the volume behind obstructions to aid mucus clearance.[6–9,34] These key mechanisms are claimed to support a PEP mask therapy for improving peripheral ventilation, which can be measured using N_2MBW test. The use of N_2MBW derivatives as a study endpoint, particularly S_{acin}*VT, assumes that the temporary increase in FRC during PEP breathing is therapeutic when delivered at 10–15 cmH_2O, thus affecting peripheral ventilation. The transitory effect of PEP therapy on FRC has been documented by few methodologically heterogenous studies, however.[35–40] The increase in FRC may not be immediate but rather occurs gradually at a subjective rate and extent that depend on lung damage severity. Taken together, the unclear albeit likely contributory role of collateral ventilation, the recruitment of collapsed airways, and the displacement of an equal pressure point induce theoretical modifications that cannot be easily measured in real time. This phenomenon could explain the unexpected S_{acin}*VT compared with the lung model theorized so far.[41,42]

Previous studies reporting differences between airway clearance interventions are difficult to interpret because of differences in endpoints, statistical approach, and study design. Very few studies remain after excluding those investigating the efficacy of ACTs as measured with sputum amount or expiratory flow, which are neither very sensitive nor specific outcomes.[43–45]

In their study, Fuchs et al.[13] used MBW based on 4% sulfur hexafluoride tracer gas and found that physiotherapy had no effect on LCI in 32 cwCF. The clinical utility of the findings is vague, since patients received 30 min of physiotherapy, which included endurance training, thorax mobilization, and ACTs with two different devices, thus being impossible to differentiate the unique contribution of each therapy on LCI. An observational study published in 2013 involved 25 cwCF aged 11.3 (3.5) years[16] who underwent a single breath washout test based on double-tracer gas to determine changes from baseline in several lung function measures, including ventilation inhomogeneity near the acinar lung regions. It is difficult to compare these findings with ours due to the lack of control, the presence of confounders, and the metrics derived from an MBW technique and device different from the ones we used.

Pfleger et al. sought more information on the effect of physiotherapy on ventilation inhomogeneity.[14] The study sample was 29 patients with CF (age range: 7.3–43.7 years) enrolled in a pre–post study that assessed changes in LCI following bronchodilator

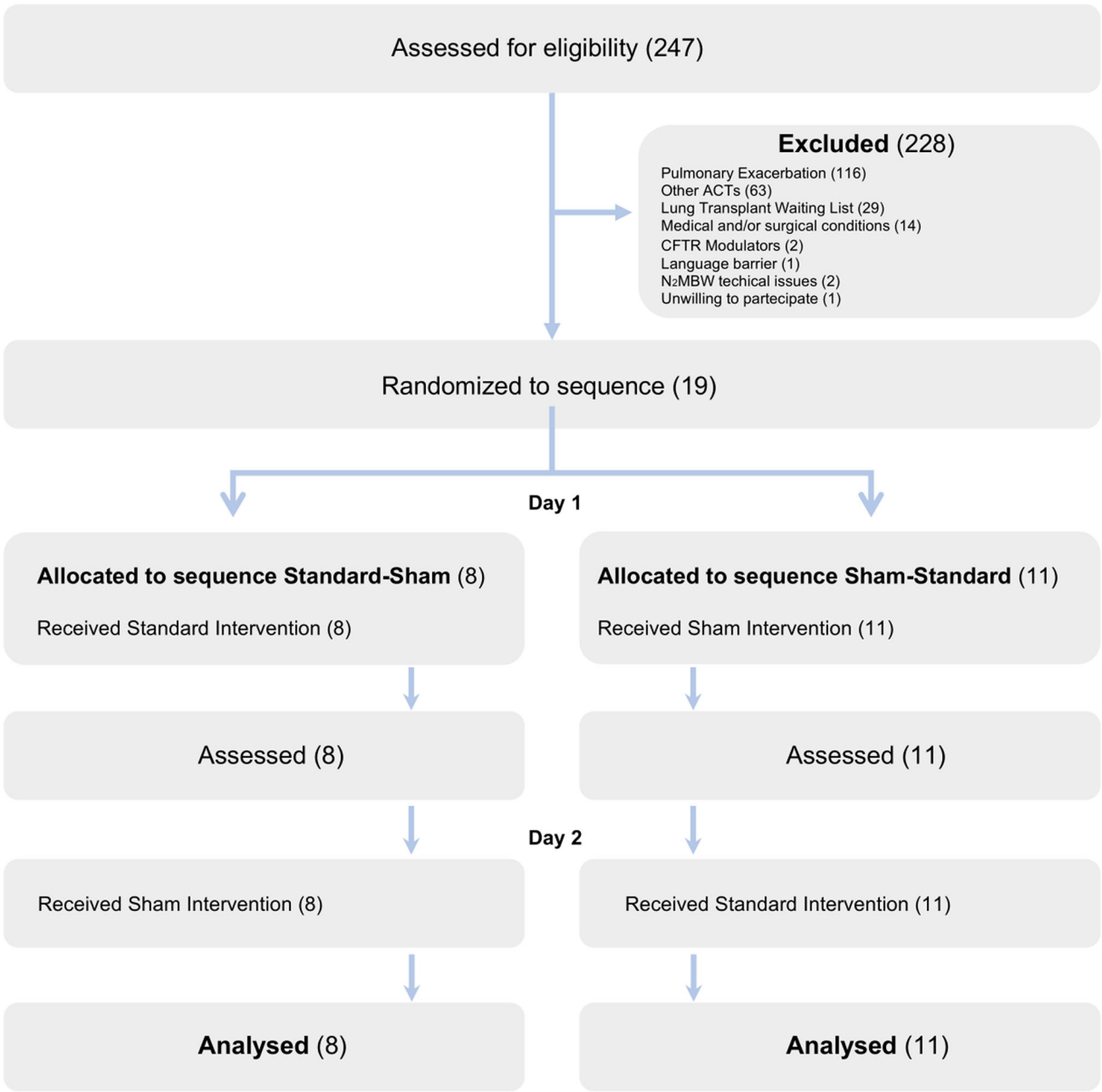

FIGURE 2 Study flow diagram. ACTs, airway clearance techniques; CFTR, cystic fibrosis transmembrane conductance regulator; N_2MBW, nitrogen multiple-breath washout. [Color figure can be viewed at wileyonlinelibrary.com]

inhalation and 30 min of PEP mask therapy. Again, the effect of physiotherapy compared with that of inhaled bronchodilation on ventilation inhomogeneity could not be properly identified, and no information about PEP mask therapy was reported. A nonrandomized study investigated the effect of physiotherapy in eight school children with CF assessed at three visits 1 month apart.[15] The assessment entailed 10 min of treadmill walking, PEP mask therapy (pressure range 10–20 cmH_2O in blocks of 15 breaths), followed by a short session of coughing and forced expiration techniques. Clinical meaningful LCI changes ≥1 were observed in only four out of the 24 measurements recorded. No conclusions could be drawn about the effect of PEP breathing on derivatives of the N_2MBW test. Eventually, a recent crossover study randomized 17 cwCF to receive PEP mask therapy delivered at 15 cmH_20 for 20 breaths, followed by a minimum of three huffs and expectoration, plus manual chest compression and 30 min rest, which was the control intervention. The study subjects crossed over to the other study arm at 6 months. No short-term effect on the derivatives of the N_2MBW test was observed; furthermore, physiotherapy was not expected to impact on the acinar airways and so was not reported.

So far, studies investigating PEP mask therapy lack internal and statistical validity, and randomized trials with a placebo and a control

TABLE 1 Baseline characteristics of study population by sequence and by total ($N = 19$).

	Treatment sequence		
	Standard-sham ($n = 8$)	Sham-standard ($n = 11$)	Total ($n = 19$)
Age (years)	11.7 (2.0)	11.2 (3.2)	11.4 (2.7)
Sex			
Males	3 (37.5)	6 (54.5)	9 (47.4)
Females	5 (62.5)	5 (45.5)	10 (52.6)
CFTR genotype			
F508del/F508del	4 (50.0)	4 (36.4)	8 (42.1)
F508del/other	3 (37.5)	4 (36.4)	7 (36.8)
other/other	1 (12.5)	3 (27.3)	4 (21.1)
BMI (*Z* score)	−1.1 (0.9)	−0.8 (0.5)	−0.9 (0.7)
Pancreatic insufficiency	8 (100.0)	10 (90.9)	18 (94.7)
FEV_1 (*Z* score)	−2.4 (1.9)	−1.2 (1.9)	−1.7 (1.9)
FRC (L)	1.4 (0.4)	1.4 (0.3)	1.4 (0.3)
S_{acin}*V (L^{-1})	0.315 (0.225)	0.187 (0.134)	0.241 (0.184)
S_{cond}*VT (L^{-1})	0.088 (0.035)	0.081 (0.030)	0.083 (0.032)
LCI	14.05 (4.27)	9.91 (2.44)	11.74 (3.8)
Pseudomonas aeruginosa infection	4 (50.0)	6 (54.5)	10 (90.9)
Inhalation therapy			
SABA	7 (87.5)	10 (90.9)	17 (89.5)
LABA + ICS	5 (62.5)	5 (45.5)	10 (52.6)
Hypertonic saline 7%	4 (50.0)	7 (63.6)	11 (57.9)
rhDNase	7 (87.5)	8 (72.7)	15 (78.9)
Levofloxacin	-	1 (9.1)	1 (5.3)
Aztreonam	1 (12.5)	-	1 (5.3)
Colistimethate sodium	-	1 (9.1)	1 (5.3)
Tobramycin	-	1 (9.1)	1 (5.3)

Note: Data are presented as mean and ±1 standard deviation (SD) or count and percentage.

Abbreviations: BMI, body mass index; CFTR, cystic fibrosis transmembrane conductance regulator; FEV_1, forced expiratory volume in the first second; FRC, functional residual capacity; ICS, inhaled corticosteroid; LABA, long-acting $beta_2$-agonist; LCI, lung clearance index; SABA, short-acting $beta_2$-agonist; S_{acin}*VT, ventilation inhomogeneity in acinar airways; S_{cond}*VT, ventilation inhomogeneity in conductive airways.

group are, therefore, needed. Defining a placebo group is challenging in physiotherapy, however, because it is difficult to simulate the active procedure, except for its therapeutic component. In addition, the behavioral and neuropsychological modifications induced by physiotherapy require making account for a possible placebo/nocebo effect in respiratory medicine.[46] Due to the rituality of physiotherapy[47] and the expected benefit of PEP mask therapy in CF, which our CF centers prescribe starting from diagnosis of the disease, withholding treatment might have aroused negative expectations, with a negative influence on study outcomes, at least in theory. For this reason, we opted for a sham-controlled study design. The sham procedure closely mimicked actual PEP mask therapy, including contextual and patient features such as monitoring of PEP breathing by a physiotherapist and the use of the same PEP device and set of resistors. Sham procedures have generally been considered more appropriate than no intervention for evaluating trial efficacy since they can more clearly determine whether a treatment is effective beyond the placebo response, which is elicited by contextual cues.[48]

In evidenced-based physiotherapy, evidence for the physiologic effects of PEP mask therapy remains scarce. When patients require personalized interventions that have been proven effective, research into airway clearance in CF is often presented as a black box of techniques carried out in nonstandardized fashion. Since PEP breathing does not consistently affect peripheral ventilation inhomogeneity in cwCF with very little secretions after hospital admission, more guidance is needed to continue prescribing PEP mask therapy in people with CF who take CFTR

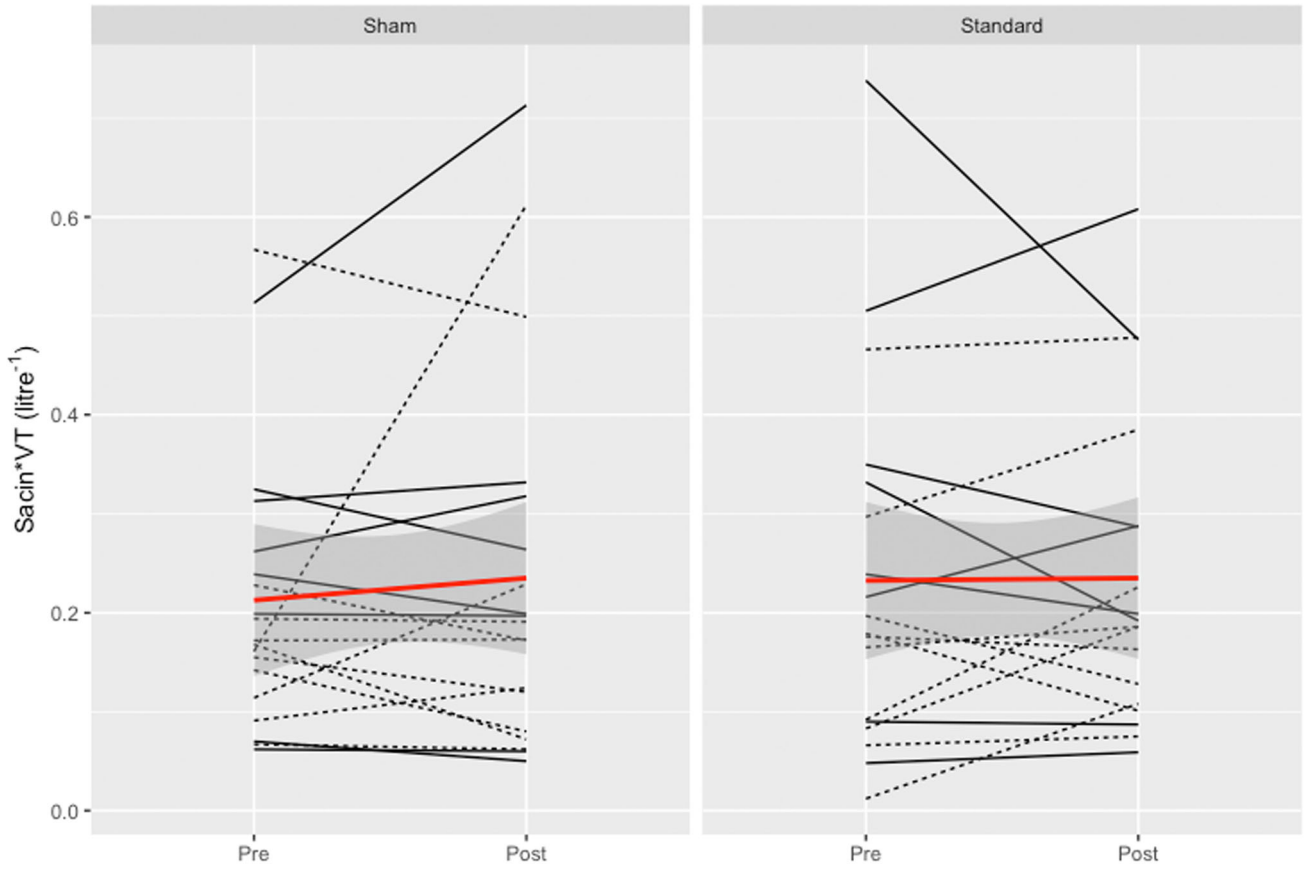

FIGURE 3 Ventilation inhomogeneity in conductive airways (S_{acin}*VT) profile for children with cystic fibrosis before and after the standard and the sham intervention. The dotted lines denote the sham-standard sequence and the continuous lines denote the standard-sham sequence. The red line denotes a regression line with 95% confidence interval as gray bands. The largest difference (0.451 L^{-1}) was recorded in a 5.6-year-old boy. [Color figure can be viewed at wileyonlinelibrary.com]

modulators, no longer present with secretion, and have better FEV_1 and LCI.[49,50] Our sensitivity analysis of the effect of PEP breathing on S_{acin}*VT based on the level of FEV_1 impairment showed no evidence of difference between the procedures.

What remains necessary is to document the physiologic effects of PEP mask therapy for planning physiotherapy in cwCF with FEV_1 < LLN who cannot access CFTR modulators.[51]

4.1 | Strength and limitations

The present study has several limitations. Since randomized controlled trials lack external validity, our findings are not easily translated into clinical settings where respiratory physiotherapy is a personalized intervention. Since we included cwCF close to their hospital discharge date, the short-term effects of PEP mask therapy might be less than expected because of the patients' relatively good condition and absence of secretions. In addition, physiotherapy is usually preceded by bronchodilator inhalation. The study conditions we set up do not reflect the real-world routine of aerosol therapy followed by airway clearance sessions.

Moreover, performing the N_2MBW test in this crossover study demanded dedicated staffing, which was an issue considering that staffing level at the two CF centers was below two full-time physiotherapists per 50 pediatric patients[52]; these aspects need to be taken into account when planning future studies investigating the short-term effects of ACTs. Also, the Milan study center reached 32% of the target sample size in 2 years of recruiting, after which the Florence center joined the trial and the study protocol was amended accordingly. This was necessitated by the rapid changes in pediatric CF care following the introduction of CFTR modulators, which has reduced scheduled hospitalization rates compared with previous years. On the whole, the study took almost five calendar years to be completed at two large CF clinics.

Furthermore, exposure to two N_2MBW tests on two consecutive mornings could be perceived as stressful for cwCF, despite a familiar and relaxing testing environment. Future research should explore other outcomes to evaluate the physiological effect of PEP mask therapy in cwCF, while keeping in mind that the delivered pressure targets the lung periphery and not the central or the proximal airway. These limitations notwithstanding, the inclusion criteria and the trial design selected to

TABLE 2 Primary and secondary outcomes.

	Standard			Sham			
	Pre	Post	Mean change (95% CI)[a]	Pre	Post	Mean change (95% CI)[a]	Effect (95% CI)[b]
Primary outcome							
Sacin*VT (L^{-1})	0.233 (0.183)	0.24 (0.149)	0.008 (−0.038 to 0.053)	0.213 (0.139)	0.235 (0.189)	0.022 (−0.038 to 0.082)	−0.015 (−∞ to 0.025)[c]
Secondary outcomes							
Scond*VT (L^{-1})	0.08 (0.045)	0.082 (0.035)	0.002 (−0.013 to 0.017)	0.091 (0.028)	0.093 (0.028)	0.002 (−0.007 to 0.011)	−0.005 (−0.019 to 0.01)
LCI	11.74 (3.8)	11.83 (3.83)	0.09 (−0.32 to 0.50)	11.80 (4.07)	11.44 (3.8)	−0.36 (−0.71 to 0.01)	0.49 (−0.05 to 1.03)

Note: Data are reported as mean (±1 SD).

Abbreviation: CI, confidence interval; LCI, lung clearance index; S_{acin}*VT, ventilation inhomogeneity in acinar airways; S_{cond}*VT, ventilation inhomogeneity in conductive airways.

[a]Unadjusted mean Post-Pre difference with 95% CI.

[b]Adjusted difference between Standard-Sham treatment with 95% CI.

[c]90% left tail CI.

maximize the effect of a single airway clearance intervention with PEP mask therapy make this a unique contribution to the current literature.

5 | CONCLUSIONS

Uncertainty surrounds the size of the short-term treatment effect in clinical trials about PEP mask therapy in CF. The findings from this study investigating the specific and the nonspecific components of PEP breathing do not provide evidence for an immediate effect of PEP mask therapy on S_{acin}*VT reduction at a pressure range of 10–15 cmH_20 in cwCF.

AUTHOR CONTRIBUTIONS

Simone Gambazza: Conceptualization; methodology; formal analysis; visualization; writing—original draft. **Alessandra Mariani**: Validation; investigation; data curation; visualization; writing—original draft. **Riccardo Guarise**: Conceptualization; methodology; investigation; writing—review and editing. **Beatrice Ferrari**: Investigation; resources; supervision; project administration; writing—review and editing. **Federica Carta**: Investigation; data curation; validation; writing—review and editing. **Anna Brivio**: Conceptualization; investigation; supervision; writing—review and editing. **Sofia Bizzarri**: Investigation; data curation; writing—review and editing. **Chiara Castellani**: Investigation; data curation; writing - review and editing. **Carla Colombo**: Supervision; resources; writing—review and editing. **Dario Laquintana**: Supervision; funding acquisition; resources; writing—review and editing.

ACKNOWLEDGMENTS

We thank the children and their families for participating in the study. Thanks are due to Stefano Becchiati for his assistance in an early stage of the study. This study was partially funded by the Italian Ministry of Health—Current research IRCCS. Open access funding provided by BIBLIOSAN.

CONFLICT OF INTEREST STATEMENT

The authors declare that the study was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest. S. G. received fees as a consultant for BGP Products Operations GmbH (Switzerland); A. B. received support from Neupharma S.r.l (Bologna, Italia) for attending a conference; RG received fees as a consultant for Vertex Pharmaceuticals S.r.l. (Italy).

DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this study will be made available by the corresponding author on request.

ETHICS STATEMENT

The study involving human subjects was reviewed and approved by the Ethics Committee Milano Area B. Written informed consent to

participate in the study was obtained from the subjects or their legal guardian/next of kin.

ORCID

Simone Gambazza http://orcid.org/0000-0002-6225-2989
Carla Colombo http://orcid.org/0000-0002-8975-695X

REFERENCES

1. Castellani C, Duff AJA, Bell SC, et al. ECFS best practice guidelines: the 2018 revision. *J Cyst Fibros*. 2018;17(2):153-178.
2. Gambazza S, Carta F, Brivio A, Colombo C. Aerosol delivery practice in Italian cystic fibrosis centres: a national survey. *Arch Physiother*. 2016;6(1):1.
3. McIlwaine M, Button B, Nevitt SJ. Positive expiratory pressure physiotherapy for airway clearance in people with cystic fibrosis. *Cochrane Database Syst Rev*. 2019;2019(11):CD003147.
4. Main E, Prasad A, Schans C. Conventional chest physiotherapy compared to other airway clearance techniques for cystic fibrosis. *Cochrane Database Syst Rev*. 2005;1:CD002011.
5. McIlwaine MP, Alarie N, Davidson GF, et al. Long-term multicentre randomised controlled study of high frequency chest wall oscillation versus positive expiratory pressure mask in cystic fibrosis. *Thorax*. 2013;68(8):746-751.
6. Groth S, Stafanger G, Dirksen H, Andersen JB, Falk M, Kelstrup M. Positive expiratory pressure (PEP-mask) physiotherapy improves ventilation and reduces volume of trapped gas in cystic fibrosis. *Bull Eur Physiopathol Respir*. 1985;21(4):339-343.
7. Martin HB. Respiratory bronchioles as the pathway for collateral ventilation. *J Appl Physiol*. 1966;21(5):1443-1447.
8. Oberwaldner B. Physiotherapy for airway clearance in paediatrics. *Eur Respir J*. 2000;15(1):196-204.
9. Oberwaldner B, Evans JC, Zach MS. Forced expirations against a variable resistance: a new chest physiotherapy method in cystic fibrosis. *Pediatr Pulmonol*. 1986;2(6):358-367.
10. Stanojevic S, Davis SD, Retsch-Bogart G, et al. Progression of lung disease in preschool patients with cystic fibrosis. *Am J Respir Crit Care Med*. 2017;195(9):1216-1225.
11. Stanojevic S, Davis SD, Perrem L, et al. Determinants of lung disease progression measured by lung clearance index in children with cystic fibrosis. *Eur Respir J*. 2021;58:2003380.
12. Vandervoort B, de Beuckeleer D, Huenaerts E, et al. The short term influence of chest physiotherapy on lung function parameters in children with cystic fibrosis and primary ciliary dyskinesia. *Front Pediatr*. 2022;10:858410.
13. Fuchs SI, Toussaint S, Edlhaimb B, Ballmann M, Gappa M. Short-term effect of physiotherapy on variability of the lung clearance index in children with cystic fibrosis. *Pediatr Pulmonol*. 2010;45(3):301-306.
14. Pfleger A, Steinbacher M, Schwantzer G, Weinhandl E, Wagner M, Eber E. Short-term effects of physiotherapy on ventilation inhomogeneity in cystic fi brosis patients with a wide range of lung disease severity. *J Cyst Fibros*. 2015;14:627-631.
15. Voldby C, Green K, Rosthøj S, et al. The effect of time-of-day and chest physiotherapy on multiple breath washout measures in children with clinically stable cystic fibrosis. *PLoS One*. 2018;13(1):e0190894.
16. Abbas C, Singer F, Yammine S, Casaulta C, Latzin P. Treatment response of airway clearance assessed by single-breath washout in children with cystic fibrosis. *J Cyst Fibros*. 2013;12(6):567-574.
17. Roethlisberger K, Nyilas S, Riedel T, Wolfensberger J, Singer F, Latzin P. Short-Term effects of elastic chest wall restriction on pulmonary function in children with cystic fibrosis. *Respiration*. 2018;96(6):535-542.
18. Stylemans D, Darquenne C, Schuermans D, Verbanck S, Vanderhelst E. Peripheral lung effect of elexacaftor/tezacaftor/ivacaftor in adult cystic fibrosis. *J Cyst Fibros*. 2022;21(1):160-163.
19. Dwan K, Li T, Altman DG, Elbourne D. CONSORT 2010 statement: extension to randomised crossover trials. *BMJ*. 2019;366:l4378.
20. Fuchs HJ, Borowitz DS, Christiansen DH, et al. Effect of aerosolized recombinant human DNase on exacerbations of respiratory symptoms and on pulmonary function in patients with cystic fibrosis. *N Engl J Med*. 1994;331(10):637-642.
21. Graham BL, Steenbruggen I, Miller MR, et al. Standardization of spirometry 2019 update an official American Thoracic Society and European Respiratory Society technical statement. *Am J Respir Crit Care Med*. 2019;200(8):e70-e88.
22. Quanjer PH, Stanojevic S, Cole TJ, et al. Multi-ethnic reference values for spirometry for the 3–95-yr age range: the global lung function 2012 equations. *Eur Respir J*. 2012;40(6):1324-1343.
23. Jensen R, Green K, Gustafsson P, et al. *Standard Operating Procedure: Multiple Breath Nitrogen Washout*. 2013. Available at https://www.mbwtraining.com/ECFS_MBW_SOP.pdf
24. Robinson PD, Latzin P, Verbanck S, et al. Consensus statement for inert gas washout measurement using multiple- and single-breath tests. *Eur Respir J*. 2013;41(3):507-522.
25. Robinson PD, Latzin P, Ramsey KA, et al. Preschool multiple-breath washout testing. an official American Thoracic Society Technical Statement. *Am J Respir Crit Care Med*. 2018;197(5):e1-e19.
26. Jensen R, Stanojevic S, Klingel M, et al. A systematic approach to multiple breath nitrogen washout test quality. *PLoS One*. 2016;11(6):e0157523.
27. Salamon ER, Gain KR, Hall GL. Defining the appropriate waiting time between multiple-breath nitrogen washout measurements. *Eur Respir J*. 2015;45(5):1489-1491.
28. Hardaker KM, Gustafsson P, Cooper P, Fitzgerald D, Selvadurai H, Robinson PD. Is twice the duration of washout sufficient time between multiple breath nitrogen washout tests? *Eur Respir J*. 2017;49(2):1501832.
29. Mortensen J, Falk M, Groth S, Jensen C. The effects of postural drainage and positive expiratory pressure physiotherapy on tracheobronchial clearance in cystic fibrosis. *Chest*. 1991;100:1350-1357.
30. Falk M, Kelstrup M, Andersen JB, et al. Improving the ketchup bottle method with positive expiratory pressure, PEP, in cystic fibrosis. *Eur J Respir Dis*. 1984;65:423-432.
31. Senn S. *Cross-Over Trials in Clinical Research*. Vol 156, 2nd ed. John Wiley & Sons, Ltd; 2002:512. doi:10.1002/0470854596
32. R Core Team. *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing; 2023. https://www.R-project.org
33. Miller FG, Rosenstein DL. Reporting of ethical issues in publications of medical research. *Lancet*. 2002;360(9342):1326-1328.
34. Zach MS, Oberwaldner B. Effect of positive expiratory pressure breathing in patients with cystic fibrosis. *Thorax*. 1992;47:66-67.
35. Bianchi R, Gigliotti F, Romagnoli I, et al. Chest wall kinematics and breathlessness during pursed-lip breathing in patients with COPD. *Chest*. 2004;125(2):459-465.
36. Garrard CS, Shah M. The effects of expiratory positive airway pressure on functional residual capacity in normal subjects. *Crit Care Med*. 1978;6(5):320-322.
37. Fagevik Olsén M, Lannefors L, Westerdahl E. Positive expiratory pressure—common clinical applications and physiological effects. *Respir Med*. 2015;109(3):297-307.
38. Tobin MJ, Chadha TS, Jenouri G, Birch SJ, Gazeroglu HB, Sackner MA. Breathing patterns. *Chest*. 1983;84(3):286-294.
39. Lumb AB, Thomas CR. *Nunn and Lumb's Applied Respiratory Physiology*. Elsevier; 2020.

40. Andersen JB, Qvist J, Kann T. Recruiting collapsed lung through collateral channels with positive end-expiratory pressure. *Scand J Respir Dis*. 1979;60(5):260-266.
41. Crawford AB, Makowska M, Paiva M, Engel LA. Convection- and diffusion-dependent ventilation maldistribution in normal subjects. *J Appl Physiol*. 1985;59(3):838-846.
42. Verbanck S, Paiva M. Model simulations of gas mixing and ventilation distribution in the human lung. *J Appl Physiol*. 1990;69(6):2269-2279.
43. Gambazza S, Zuffo S. CPAP in cystic fibrosis: is it time to surrender yet? *Respir Care*. 2013;58(9):e116-e117.
44. Marques A, Bruton A, Barney A. Clinically useful outcome measures for physiotherapy airway clearance techniques: a review. *Phys. Ther. Rev*. 2006;11(4):299-307.
45. Rubin BK. Designing clinical trials to evaluate mucus clearance therapy. *Respir Care*. 2007;52(10):1348-1358; discussion 1358-1361.
46. Meissner K. Placebo responses on cardiovascular, gastrointestinal, and respiratory organ functions. In Benedetti, F., Enck, P., Frisaldi, E., Schedlowski, M. eds. *Placebo. Handbook of Experimental Pharmacology*. Vol 225. Springer; 2014:183-203. doi:10.1007/978-3-662-44519-8_11
47. Testa M, Rossettini G. Enhance placebo, avoid nocebo: how contextual factors affect physiotherapy outcomes. *Man Ther*. 2016;24:65-74.
48. Brim RL, Miller FG. The potential benefit of the placebo effect in sham-controlled trials: implications for risk-benefit assessments and informed consent. *J Med Ethics*. 2013;39:703-707.
49. Graeber SY, Renz DM, Stahl M, et al. Effects of elexacaftor/tezacaftor/ivacaftor therapy on lung clearance index and magnetic resonance imaging in patients with cystic fibrosis and one or two F508del alleles. *Am J Respir Crit Care Med*. 2022;206(3):311-320.
50. Graeber SY, Boutin S, Wielpütz MO, et al. Effects of lumacaftor-ivacaftor on lung clearance index, magnetic resonance imaging, and airway microbiome in Phe508del homozygous patients with cystic fibrosis. *Ann Am Thorac Soc*. 2021;18(6):971-980.
51. Guo J, Garratt A, Hill A. Worldwide rates of diagnosis and effective treatment for cystic fibrosis. *J Cyst Fibros*. 2022;21(3):456-462.
52. Conway S, Balfour-Lynn IM, De Rijcke K, et al. European cystic fibrosis society standards of care: framework for the cystic fibrosis centre. *J Cyst Fibros*. 2014;13(S1):S3-S22.

How to cite this article: Gambazza S, Mariani A, Guarise R, et al. Short-term effects of positive expiratory pressure mask on ventilation inhomogeneity in children with cystic fibrosis: a randomized, sham-controlled crossover study. *Pediatr Pulmonol*. 2024;1-10. doi:10.1002/ppul.26915