Diaphragm dysfunction in critically ill patients
from monitoring to interventions

Jonne Doorduin
Diaphragm dysfunction in critically ill patients
from monitoring to interventions

Jonne Doorduin
Diaphragm dysfunction in critically ill patients, from monitoring to interventions

ISBN: 978-90-826784-2-0

The work described in this thesis was performed at the Department of Intensive Care Medicine and Radboud Institute for Health Sciences, Radboud university medical center, Nijmegen.

Cover design, lay-out and printing: proefschrift-aio.nl

Financial support for publication of this thesis was kindly provided by: Radboud University Nijmegen, Maquet Netherlands B.V. and Twente Medical Systems International B.V.

©2017 Jonne Doorduin
All rights reserved. No part of this thesis may be reproduced or transmitted in any form or by any means, electronic or mechanical, including photocopy, recording or otherwise without permission of the author.
Diaphragm dysfunction in critically ill patients
from monitoring to interventions

Proefschrift

ter verkrijging van de graad van doctor
aan de Radboud Universiteit Nijmegen
op gezag van de rector magnificus prof. dr. J.H.J.M. van Krieken,
volgens besluit van het college van decanen
in het openbaar te verdedigen op donderdag 22 juni 2017
om 12.30 uur precies

door

Jonne Doorduin
geboren op 14 januari 1985
te Gieten
**Promotoren**
Prof. dr. J.G. van der Hoeven
Prof. dr. L.M.A. Heunks (VU medisch centrum)

**Copromotor**
Dr. H.W.H. van Hees

**Manuscriptcommissie**
Prof. dr. G.J. Scheffer
Prof. dr. M.T.E. Hopman
Prof. dr. ir. M.J.A.M. van Putten (Universiteit Twente)
Diaphragm dysfunction in critically ill patients from monitoring to interventions

Doctoral Thesis

to obtain the degree of doctor from Radboud University Nijmegen on the authority of the Rector Magnificus prof. dr. J.H.J.M. van Krieken, according to the decision of the Council of Deans to be defended in public on Thursday, June 22, 2017 at 12.30 hours

by

Jonne Doorduin
Born on January 14, 1985 in Gieten (the Netherlands)
Supervisors
Prof. dr. J.G. van der Hoeven
Prof. dr. L.M.A. Heunks (VU University Medical Center)

Co-supervisor
Dr. H.W.H. van Hees

Doctoral Thesis Committee
Prof. dr. G.J. Scheffer
Prof. dr. M.T.E. Hopman
Prof. dr. ir. M.J.A.M. van Putten (University of Twente)
## Contents

**Chapter 1**  
Introduction and outline of the thesis  

**Part I  Monitoring**

**Chapter 2**  
Monitoring of the respiratory muscles in the critically ill  

**Chapter 3**  
The differential diagnosis for failure to wean from mechanical ventilation  
*Curr Opin Anaesthesiol. 2016 Apr;29(2):150-7*

**Chapter 4**  
Inspiratory and expiratory muscle effort during successful and failed weaning from mechanical ventilation  
*Submitted for publication*

**Chapter 5**  
Functional assessment of the diaphragm by ultrasonic deformation imaging during inspiratory loading  
*Accepted for publication in J Appl Physiol*

**Part II  Interventions**

**Chapter 6**  
Automated patient-ventilator interaction analysis during non-invasive NAVA and pressure support ventilation in patients with COPD  

**Chapter 7**  
Assisted ventilation in patients with acute respiratory distress syndrome: lung-distending pressure and patient-ventilator interaction  
*Anesthesiology. 2015 Jul; 123(1):181-90*
Chapter 8  Partial neuromuscular blocking during partial ventilator support in sedated patients with high tidal volumes
*Am J Resp Crit Care Med. 2017 Apr 15;195(8):1033-1042*

Chapter 9  The calcium sensitizer levosimendan improves human diaphragm function

Chapter 10  Effects of experimental human endotoxemia on diaphragm function
*Shock. 2015 Oct;44(4):316-22*

Chapter 11  Summary, general discussion and future perspectives

Chapter 12  Nederlandse samenvatting, algehele discussie en toekomstperspectieven

Dankwoord
List of publications
Curriculum Vitae
Chapter 1

Introduction and outline of the thesis
Introduction and outline of the thesis

Critically ill patients
Critical illness can be defined as a life-threatening condition caused by marked alterations in the structure and function of vital organs that can result in significant morbidity and mortality. In the Netherlands, each year nearly 100,000 patients are admitted to the intensive care unit (ICU) with a mortality rate of 15-20% (1, 2). Severe infections remain the most common problem encountered on the ICU (1), but also surgery and trauma can lead to critical illness. To save lives, continuous monitoring of vital organ functions in critically ill patients is crucial in order to timely start and evaluate therapeutic interventions. For example, sophisticated monitoring techniques in patients with acute respiratory failure on mechanical ventilation may limit development of complications associated with mechanical ventilation, such as lung injury and diaphragm dysfunction.

The diaphragm
The diaphragm is our main respiratory muscle and accounts for the major part (approximately 70%) of inspiratory work (3), hence it is called the respiratory pump. The diaphragm is a dome-shaped, musculotendinous partition separating the thoracic and abdominal cavities (Figure 1). The diaphragm is a skeletal muscle and receives its motor innervation via the phrenic nerve, which originates from the cervical roots 3 to 5. During inspiration the diaphragm descends; however, only its central part moves because its periphery, as the fixed origin of the muscle, attaches to the inferior margin of the thoracic cage and the superior lumbar vertebrae.

During inspiration, the downward movement of the diaphragm and outward movement of the chest wall results in a decrease in pleural pressure, i.e. the pressure surrounding the lung within the pleural space. Consequently pressure in the alveoli decreases, which creates a pressure gradient between the airway opening and the alveoli, leading to flow of air into the lungs. The transpulmonary pressure is the pressure difference between the opening to the pulmonary airway and the pleural surface, it represents the distending pressure of the lungs (4). In order to determine the contribution of the diaphragm to breathing, one can use the abdominal pressure. Due to the downward movement of the diaphragm, abdominal pressure increases during inspiration. When diaphragm function is impaired, the magnitude of increase in abdominal pressure is reduced and may even decrease during inspiration, this is clinically referred to as paradoxical chest movement. The pressure gradient over the diaphragm can therefore be estimated by subtracting pleural pressure from abdominal
Introduction and outline of the thesis

1

pressure. Gastric pressure (Pga) may represent abdominal pressure. Unfortunately, pressures within the chest cavity are difficult to measure and an estimate of pleural pressure is required. As the esophagus is situated inside the thoracic cavity, the esophageal pressure (Pes) can be used as an estimate for pleural pressure. The pressure gradient over the diaphragm can then be calculated as: transdiaphragmatic pressure (Pdi) = Pga – Pes.

Figure 1. The inner anterior surface (left) and inner posterior surface (right) of the diaphragm. Right: the three large apertures from top to bottom are the caval opening, the esophageal hiatus and the aortic hiatus. Adapted from Cluzel and colleagues (5).

Critical illness and diaphragm dysfunction

Critical illness has detrimental effects on the structure and function of the diaphragm (6-12). For instance, respiratory muscle strength measured by volitional maximal inspiratory pressure, has been shown to decrease after one week of mechanical ventilation and is associated with delayed extubation and prolonged ventilation (7). Also, non-volitional measurement of diaphragm strength, by phrenic nerve stimulation, revealed severe diaphragm weakness in critically ill patients (8). Diaphragm weakness may already be present at ICU admission and is associated with sepsis and disease severity (10, 13), suggesting it is an important part of multiorgan failure. The effects of critical illness on diaphragm function are often part of a more generalized phenomenon, known as ICU-acquired weakness (14). Factors such as systemic inflammation, drugs, electrolyte disturbances, and immobility have been identified in the pathogenesis of ICU-acquired skeletal muscle weakness (14). Besides generalized muscle weakness, there is convincing evidence that mechanical ventilation is an important cause of diaphragm dysfunction, so-called ventilator-induced diaphragm dysfunction (VIDD) (15). In a landmark paper, Levine and colleagues demonstrated that only a few days of controlled mechanical ventilation is
associated with disuse atrophy of the diaphragm (6). In accordance with these data, Jaber and colleagues reported the functional consequences of critical illness on the respiratory muscles (9). They found approximately a third reduction in respiratory muscle function induced in the first 5-6 days of invasive mechanical ventilation, indicating the rapid development of diaphragm weakness. Recently, the frequent development of diaphragm atrophy in critically ill patients was also demonstrated using diaphragm ultrasound (16). Moreover, the diaphragm is more severely affected by critical illness and mechanical ventilation than other peripheral muscles (6, 17). At the time of liberation from mechanical ventilation, diaphragm dysfunction exists twice as frequent as limb muscle weakness and is associated with subsequent weaning failure and mortality (17). Therefore, there is an urgent need for tools to monitor diaphragm function and to develop preventive and therapeutic interventions to improve diaphragm function in critically ill patients.

Mechanical ventilation and patient-ventilator interaction

For patients with acute respiratory failure mechanical ventilation is a life-saving intervention, which aims to reduce the work of breathing and improve gas exchange. Modern mechanical ventilators support or substitute the inspiratory effort of the diaphragm by delivering air to the lungs via positive pressure, causing an increase in pleural pressure. The latter is in contrast to unassisted spontaneous breathing, where caudal movement of the diaphragm causes a decrease in pleural pressure. Positive pressure ventilation can be delivered to the patient via an invasive or non-invasive route. Invasive mechanical ventilation requires placement of an endotracheal tube, whereas non-invasive mechanical ventilation is delivered via a face mask. Positive pressure ventilation is usually delivered with a bi-level approach. A lower pressure level during the expiratory phase, called positive end-expiratory pressure, and a higher level of assist during the inspiratory phase. Ventilator assist can be provided in a ‘controlled’ or ‘assisted’ mode. With controlled ventilation, duration and frequency of the inspiratory phase is completely regulated by the ventilator. With assisted ventilation, patients are required to trigger the ventilator, allowing the patient to time assist delivery to inspiratory effort. Triggering in conventional ventilators takes place by detection of the inspiratory flow or pressure generated by the patient and cycling-off is flow-based.

To avoid patients ‘fighting the ventilator’ with assisted ventilation, the interaction between patient and ventilator is important. Ideally, the level of assist should be delivered in proportion to respiratory effort of the patient and in perfect synchrony with patient neural inspiratory time and neural expiratory time. Too high levels of inspiratory support may suppress output from the respiratory centers (over-assist),
which is associated with respiratory muscle atrophy and contractile dysfunction (6, 16, 18). In opposite, too low levels of assist may result in high work of breathing and strenuous diaphragm efforts, which may result in the development of diaphragm injury and fatigue (19-21). In practice delivering synchronous and proportional assist is challenging with flow or pressure controlled systems (22). Patient-ventilator interaction depends on factors related to the ventilator and the patient. Ventilator-related factors are the trigger sensitivity, the variable that control gas delivery, and the cycling-off criterion (23). Patient-related factors include the mechanics of the respiratory system and neural activation of the diaphragm (23). Patient-ventilator interaction can be classified into synchronous, dyssynchronous or asynchronous respiratory events (24). Dyssynchronous events refer to trigger delays and early or late cycling-off. Asynchronous events refer to wasted efforts, auto-triggering, and double-triggering. One-fourth of patients exhibit a high incidence of asynchrony (defined as more than 10% asynchronous events of total respiratory counts) during invasive assisted mechanical ventilation (25). Such a high incidence is associated with a prolonged duration of mechanical ventilation (25). With non-invasive ventilation, even 43% of patients presents with severe asynchrony (26).

A relatively new assisted mode of ventilation is neurally adjusted ventilatory assist (NAVA) (27). A mode designed to optimize patient-ventilator interaction. NAVA uses the electrical activity of the diaphragm to control the ventilator. Both timing and magnitude of assist are controlled by the patient. Clinical and physiological studies are needed to evaluate this new mode of ventilation.

Weaning from mechanical ventilation
Prolonged mechanical ventilation leads to increased morbidity, including diaphragm dysfunction, and mortality in critically ill patients (28, 29). Therefore mechanical ventilation should be discontinued as soon as possible. This process is called weaning and it covers the entire process of liberating the patient from mechanical ventilation and the endotracheal tube (30). Approximately 40% of the time a patient is mechanically ventilated is dedicated to weaning from mechanical ventilation (31). An important stage in the weaning process is a spontaneous breathing trial (SBT). During an SBT the patient is challenged to breathe without support (a T-tube trial) for a certain time, usually between 30 and 120 min. An SBT is predictive for extubation failure or extubation success of the patient. Based on the number of SBT and the number of days in between until successful extubation, weaning patients can be classified into three groups (30). Group 1, the simple weaning group includes patient who successfully pass the initial SBT and are successfully extubated on the first attempt. This group represent the majority of weaning patients. Group 2, difficult
weaning, includes patients who require up to three SBT or as long as 7 days from
the first SBT to achieve successful weaning. Group 3, prolonged weaning, includes
patients who require more than three SBT or more than 7 days of weaning after the
first SBT. This group is estimated to be approximately 6 - 15% of weaning patients
(29, 30, 32). A major determinant of weaning failure is respiratory muscle dysfunction
(7, 17). Detailed analysis of diaphragm function during an SBT might help to identify
diaphragm dysfunction in patient who fail a trial of weaning.

Aims and outline of the thesis
This thesis focuses on diaphragm function in critically ill patients. In spite of growing
evidence that diaphragm dysfunction develops in critically ill patients and contributes
to prolonged mechanical ventilation and weaning failure, the respiratory muscles
remain poorly monitored in the ICU and almost no interventions to maintain or improve
function are available. This is in sharp contrast with the numerous options available to
monitor and treat, for example, disorders of the heart and lungs in critically ill patients.
Therefore, we investigated techniques to monitor diaphragm function, and preventive
and therapeutic interventions to optimize diaphragm function.

The first part of this thesis focuses on monitoring of diaphragm function in critically
ill patients. In chapter 2 we review the techniques available to monitor respiratory
muscle function and discuss the possible implications of monitoring respiratory muscle
function in the critically ill. If not monitored, diaphragm dysfunction often becomes
apparent when a patient fails to wean from mechanical ventilation. However, the
pathophysiology of difficult and prolonged weaning may be complex. Therefore, in
chapter 3, we discuss in depth the differential diagnosis of difficult and prolonged
weaning from mechanical ventilation, with emphasis on diaphragm dysfunction.

Analysis of respiratory muscle function is an important step that is required to
identify the primary cause for prolonged weaning from mechanical ventilation. In
chapter 4 we performed detailed physiological analysis of inspiratory muscle activity
and expiratory muscle activity during an SBT, in particular neuromechanical efficiency
of the diaphragm and expiratory muscle effort were studied. Besides physiological
measurements of the diaphragm, ultrasound could be a useful and accurate
noninvasive bedside tool to evaluate diaphragm function (33). Currently, ultrasound
of the diaphragm is used to quantify thickness, thickening during inspiration and
movement of the diaphragm (34). However, correlations between physiological
measurements, such as transdiaphragmatic pressure, and diaphragm thickening are
low (35). Therefore in chapter 5 we evaluated the performance of speckle tracking
imaging to quantify diaphragm function. Speckle tracking imaging is an ultrasound
technique which enables angle-independent, two-dimensional quantification of muscle deformation and muscle deformation velocity during muscle contraction.

The second part of this thesis focuses on preventive and therapeutic interventions to optimize diaphragm function. A first approach is to prevent the development of VIDD by minimizing the harmful effects of mechanical ventilation on diaphragm function. Ideally, mechanical ventilation should exactly provide ventilatory assist proportional to the demands of the patient and cycle in perfect synchrony with neural respiratory timing of the patient. With conventional pressure support ventilation it can be challenging to deliver support in synchrony with the patients neural respiratory drive (22). NAVA is relatively new ventilator mode, controlled by electrical activity of diaphragm, where the patient regulates both timing and magnitude of assist (27). In chapter 6, we evaluated patient-ventilator interaction during different modes of non-invasive ventilation including NAVA. Previously it was shown in a heterogeneous groups of critically ill patients that non-invasive NAVA improves patient-ventilator interaction. Therefore, our study was performed in patients with chronic obstructive pulmonary disease as these patients are more likely to exhibit severe patient ventilator asynchrony (36). In addition, we used an automated analysis method to quantify patient-ventilator interaction.

In patients with acute respiratory distress syndrome (ARDS) the use of assisted mechanical ventilation over controlled mechanical ventilation is subject of debate (37-40). Assisted ventilation better preserves diaphragm function compared to controlled ventilation, but allowing spontaneous breathing efforts increases the risk of lung injury. It has been shown in patients with acute respiratory failure that increasing levels of support with NAVA unloads the respiratory muscles, but limits pressure increases due to down-regulation of respiratory muscle activity (41-43), suggesting a long-protective feedback mechanism. In chapter 7 we evaluated whether differences in patient control of the ventilator, affect lung protective ventilation, breathing pattern variability, and patient ventilator-interaction in mild to moderate ARDS patients.

After the transition from controlled mechanical ventilation to assisted mechanical ventilation, ARDS patients might generate high tidal volumes. To prevent lung injury clinicians often switch back to controlled ventilation, including high dose sedatives. However, this promotes diaphragm dysfunction. In chapter 8 we investigated whether partial neuromuscular blocking of the diaphragm can facilitate lung protective ventilation while maintaining diaphragm activity under assisted ventilation.

Currently, there are no therapeutic agents available to improve diaphragm function. In chapter 9 we studied the effects of the calcium sensitizer levosimendan
on diaphragm function in healthy subjects. Previously, we have shown that levosimendan enhances calcium sensitivity of permeabilized muscle fibers obtained from the human diaphragm (44).

As described, sepsis is strongly associated with diaphragm dysfunction (10, 13). In chapter 10 we aimed to develop an experimental in vivo model for diaphragm dysfunction during systemic inflammation. Such a model would allow testing of therapeutic agents to improve diaphragm dysfunction.
References


Introduction and outline of the thesis
Part I
Monitoring
Chapter 2

Monitoring of the respiratory muscles in the critically ill

Jonne Doorduin, Hieronymus W.H. van Hees, Johannes G. van der Hoeven, Leo M.A. Heunks

Abstract

Evidence has accumulated that respiratory muscle dysfunction develops in critically ill patients and contributes to prolonged weaning from mechanical ventilation. Accordingly, it seems highly appropriate to monitor the respiratory muscles in these patients. Today, we are only at the beginning of routinely monitoring respiratory muscle function. Indeed, most clinicians do not evaluate respiratory muscle function in critically ill patients at all. However, practical issues and the absence of sound scientific data for clinical benefit should in our opinion not discourage clinicians for having a closer look at respiratory muscle function in critically ill patients. This perspective discusses the latest developments in the field of respiratory muscle monitoring and possible implications of monitoring respiratory muscle function in critically ill patients.
Introduction

The physiological status of critically ill patients is characterized by rapidly evolving and frequently life-threatening derangements as well as ‘silent’ yet important alterations in organ function. The intensive care unit (ICU) environment is specifically equipped to monitor the critically ill patient. Intensivists monitor cardiac-, pulmonary-, and kidney function more or less continuously, and other organs or tissues, such as bone-marrow and gastrointestinal tract are closely monitored as well.

In spite of growing evidence that respiratory muscle dysfunction develops in critically ill patients and contributes to weaning failure (1-3), the respiratory muscles are poorly monitored in the ICU. Therefore, diaphragm dysfunction is usually unrecognized in the ICU and only becomes apparent when a patient fails to wean from mechanical ventilation. This may be related to: 1) the limited knowledge of health care workers on the effects of critical illness on respiratory muscle function; 2) the limited availability and knowledge of tools to monitor respiratory muscle function in critically ill patients; or 3) the perception that monitoring respiratory muscle function has no clinical consequences.

In this perspective, we discuss the latest developments in the field of respiratory muscle monitoring (Table 1) and possible implications of monitoring respiratory muscle function in the critically ill. The effects of critical illness on respiratory muscle function will be described briefly.

What is the effect of critical illness on respiratory muscle function?

The effects of critical illness on respiratory muscle function are often part of a more generalized phenomenon, known as ‘ICU acquired weakness’. It should be recognized that ICU weakness may result from alteration in muscle or nerve function, and in fact both often migrate together. Factors such as systemic inflammation, drugs, electrolyte disturbances, and immobility have been identified in the pathogenesis of ICU-acquired weakness (4). The clinical relevance of ICU-acquired weakness is supported by the observations that prolonged mechanical ventilation is associated with decreased diaphragm strength (3), and that muscle weakness is among the most prominent long-term complications of survivors of acute respiratory distress syndrome (ARDS) (5).
### Table 1. Overview of available tools to monitor respiratory muscle function.

<table>
<thead>
<tr>
<th>diagnostic tool</th>
<th>Parameter</th>
<th>Comment</th>
</tr>
</thead>
<tbody>
<tr>
<td>pressure and flow recordings</td>
<td>inspiratory muscle</td>
<td>Easy to perform voluntary measurements of global respiratory muscle strength. High values exclude respiratory muscle weakness. Low values may reflect poor technique or effort rather than respiratory muscle weakness (6).</td>
</tr>
<tr>
<td></td>
<td>strength (P\text{Imax})</td>
<td></td>
</tr>
<tr>
<td></td>
<td>expiratory muscle</td>
<td></td>
</tr>
<tr>
<td></td>
<td>strength (P\text{Emax})</td>
<td></td>
</tr>
<tr>
<td></td>
<td>neural respiratory</td>
<td>Available in most mechanical ventilators, but of limited value due to wide normal range (7).</td>
</tr>
<tr>
<td></td>
<td>drive (P\text{0.1})</td>
<td></td>
</tr>
<tr>
<td>transdiaphragmatic pressure</td>
<td>diaphragm strength</td>
<td>Voluntary measures of specific diaphragm strength. High values exclude respiratory muscle weakness. Low values may reflect poor technique or effort rather than respiratory muscle weakness (6).</td>
</tr>
<tr>
<td>*</td>
<td>(P\text{dimax} / \text{sniff Pdi})</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Gilbert index</td>
<td>These indices are frequently used for research purposes, but without dedicated software are too complicated for routine clinical use.</td>
</tr>
<tr>
<td></td>
<td>tension time index /</td>
<td></td>
</tr>
<tr>
<td></td>
<td>pressure-time product</td>
<td></td>
</tr>
<tr>
<td>electromyography*</td>
<td>neural respiratory</td>
<td>Direct measure of respiratory output from the brainstem, no normal values available.</td>
</tr>
<tr>
<td></td>
<td>drive (E\text{Adi})</td>
<td></td>
</tr>
<tr>
<td></td>
<td>patient-ventilator</td>
<td>Gold standard for detection of patient-ventilator asynchronies.</td>
</tr>
<tr>
<td></td>
<td>synchrony</td>
<td></td>
</tr>
<tr>
<td></td>
<td>neuro-ventilatory and- mechanical efficiency</td>
<td>Relatively new indices still under evaluation, no normal values available.</td>
</tr>
<tr>
<td>phrenic nerve stimulation</td>
<td>diaphragm strength</td>
<td>Non-voluntary evaluation of diaphragm function, fairly invasive and technical difficult. Should be performed only in experienced centers in selected patients.</td>
</tr>
<tr>
<td></td>
<td>(P\text{ditw} / P\text{motw})</td>
<td></td>
</tr>
<tr>
<td>imaging</td>
<td>diaphragm position</td>
<td>Atelectasis, pneumonia and diaphragmatic eventration complicate findings of hemidiaphragmatic elevation.</td>
</tr>
<tr>
<td>chest x-ray</td>
<td></td>
<td></td>
</tr>
<tr>
<td>fluoroscopy</td>
<td>diaphragm motion</td>
<td>Misleading in patients with bilateral diaphragmatic paralysis and radiation exposure (8).</td>
</tr>
<tr>
<td>ultrasonography (B/M-mode)</td>
<td>diaphragm thickness /</td>
<td>Well characterized, noninvasive and easy to perform at bedside (9). M-Mode difficult to perform for left hemidiaphragm. Technique of limited value during assisted breathing.</td>
</tr>
<tr>
<td></td>
<td>motion</td>
<td></td>
</tr>
<tr>
<td>CT / MRI</td>
<td>diaphragm position /</td>
<td>Applicability for monitoring is very limited.</td>
</tr>
<tr>
<td></td>
<td>motion</td>
<td></td>
</tr>
<tr>
<td>circulatory biomarkers</td>
<td>troponin I</td>
<td>The relation between plasma troponin I levels and functional measures in critically ill patients has not yet been investigated.</td>
</tr>
</tbody>
</table>

Abbreviations: CT = computed tomography; E\text{Adi} = electrical activity of the diaphragm; MRI = magnetic resonance imaging; P\text{0.1} = mouth pressure generated in the first 100 ms of inspiration; P\text{di} = transdiaphragmatic pressure; PE\text{max} = maximal expiratory pressure; P\text{Imax} = maximal inspiratory pressure; P\text{mo} = mouth pressure. * Requires placement of naso-gastric tube.
Besides generalized weakness, there is evidence that mechanical ventilation itself is an important cause of diaphragm dysfunction, which is collectively referred to as 'ventilator-induced diaphragm dysfunction' (VIDD) (10). In a landmark paper, Levine and colleagues demonstrated that only a few days of controlled mechanical ventilation is associated with atrophy of the diaphragm, but not to the pectoralis major (11). In a recent study, Jaber and colleagues reported the functional consequences of critical illness on respiratory muscles (2). They found ~30% reduction in twitch airway pressure induced by magnetic phrenic nerve stimulation in the first 5-6 days of invasive mechanical ventilation, indicating the rapid development of diaphragm weakness.

The last decade the understanding of the molecular and cellular mechanisms underlying respiratory muscle weakness in the critically ill has been the subject of intensive research (extensively reviewed in (12) and (13)). To summarize, an imbalance between proteolysis and protein synthesis results in a loss of contractile proteins (11, 14). In addition, function of remaining muscle proteins may be impaired by enhanced oxidation and dephosphorylation (15-17). Inflammation and oxidative stress are the major drivers of these impairments (17). Figure 1 provides a graphic representation of conditions and their pathways contributing to acquired diaphragm weakness in critically ill patients.

How to assess respiratory muscle function in critically ill patients?

A. History, symptoms and signs
A patient’s past medical history can provide information on the patient’s premorbid functional status and, in particular, preexistent respiratory muscle dysfunction. Weakness of the respiratory muscles occur in a variety of neuromuscular disorders (18), but also in non-myopathic chronic diseases such as chronic obstructive pulmonary disease (COPD) and congestive heart failure (19, 20). Other conditions that may precipitate respiratory and peripheral muscle dysfunction include aging (sarcopenia) and cachexia (21, 22). A patient’s medication history should be reviewed, since respiratory muscle function can be negatively affected by drugs including steroids, sedatives, analgetics (23-25).

Clinical examination may reveal evidence of respiratory muscle weakness. Accessory respiratory muscle recruitment, especially the sternocleidomastoid muscle, may be apparent by palpation in patients where inspiratory load exceeds the capacity of the diaphragm (26). When the diaphragm is very weak, a supine abdominal paradox can be observed.
Figure 1. Graphic representation of conditions and their pathways contributing to acquired diaphragm weakness in critically ill patients. As shown, different conditions can lead to diaphragm atrophy via an imbalance between proteolysis and protein synthesis (11, 14), whereas remaining muscle proteins may be impaired by enhanced oxidation and dephosphorylation (15-17). Inflammation and oxidative stress are proposed to be the major drivers of these impairments (17). In addition, certain drugs can impair neural drive and excitation-contraction coupling. Abbreviations: NMBA = neuromuscular blocking agent.

Conversely, care must be taken in assessing abdominal wall motion in patients actively exhaling. Contraction of the abdominal muscles during expiration and subsequent relaxation as an assistance to inspiration may give the appearance of outward motion of the anterior abdominal wall during inspiration. Thus, activation of the abdominal muscles during expiration could also be regarded as a sign of respiratory muscle dysfunction.

B. Pressure and flow recordings
Airway pressure and flow
Although routine assessment of respiratory muscle function using airway pressures is common in several diseases, such as COPD, very few clinicians actually measure global respiratory muscle strength in the ICU. A maximal static inspiratory (Mueller maneuver) and expiratory maneuver can be obtained in intubated patients to evaluate global inspiratory ($P_{\text{Imax}}$) and expiratory ($P_{\text{Emax}}$) muscle strength. Both can be measured.
Monitoring of the respiratory muscles in the critically ill

either while the patient is connected to the ventilator or during brief disconnection using a handheld pressure monitoring device. Voluntary maneuvers require patient cooperation and are influenced by sedation level, anxiety and pain. Therefore, high values exclude clinically significant weakness, but low values are common and may reflect poor technique or effort as well (6). To obtain more reliable measurements of \( P_{\text{Imax}} \) in ventilated and sedated patients, a 20 second end-expiratory occlusion period can be performed (27). A T-piece with one-way valves should be attached to the endotracheal tube to allow expiration, but obstruct inspiration.

Airway pressure (Paw) generated in the first 100 ms of inspiration (\( P_{0.1} \)) has been used as an index of neural respiratory drive. Interpretation of \( P_{0.1} \) is limited by its wide normal range and dependency on lung volume and contractile properties of the diaphragm, nevertheless the technique remains useful when its limitations are recognized (7).

**Transdiaphragmatic pressure**

In contrast to \( P_{\text{Imax}} \), a specific measure of diaphragm muscle strength is transdiaphragmatic pressure (\( P_{\text{di}} \)). \( P_{\text{di}} \) is the difference between abdominal and pleural pressure. In practice, the difference between esophageal (Pes) and gastric pressure (Pga) is used to calculate \( P_{\text{di}} \). Voluntary measurements of maximum \( P_{\text{di}} \) can be obtained by having the patients inspire as forcefully as possible against a closed airway (28), or by having the patient sniff forcefully (29). Sniff \( P_{\text{di}} \) appears to be more reproducible than maximum inspiratory \( P_{\text{di}} \) (29). The Gilbert index (\( \Delta \text{Pga}/\Delta \text{Pdi} \)) can be used to determine the relative contribution of the diaphragm to inspiration (30). The higher this index, the greater is the contribution of the diaphragm to total inspiratory effort. In case of a paralyzed diaphragm, the Gilbert index becomes negative. To estimate the energy expenditure of the diaphragm, the tension-time index and pressure-time product of the diaphragm can be calculated using \( P_{\text{di}} \) (31, 32). These indices are frequently used for research purposes, but without dedicated software are too complicated for routine clinical use.

Today, gastric- and esophageal balloons are extensively used for research purpose but not routinely in clinical care. This is probably the result of the perceived invasiveness of the procedure and technical difficulties. It should be noted that techniques, that are much more invasive and probably at least as complex (i.e. pulmonary artery catheter) are used to measure cardiac function in selected critically ill patients. Interestingly, in a recent study, esophageal pressure measurement was used to guide PEEP setting, indicating the applicability in clinical care (33). In fact, some of the modern ventilators have auxiliary ports to measure esophageal pressure, which is a step forward to implement measurement of esophageal pressure in clinical care.
However, one should keep in mind that Pdi is influenced by positive pressure of the mechanical ventilator and ideally should be measured during a trial of spontaneous breathing.

In conclusion, \( P_{\text{Imax}} \) and \( P_{E_{\text{max}}} \) can be used as a global measure for respiratory muscle function and possibly to monitor the response of respiratory muscle training. We recommend the use of esophageal catheters only for monitoring diaphragm function in selected ICU patients, such as with difficult weaning (34). In these patients, esophageal and gastric pressures could be used to closely monitor the role of the diaphragm in weaning failure (34).

### C. Electromyography

Electromyography (EMG) comprises the temporal and spatial summation of neural impulses from the brain that are translated into muscle fiber action potentials. Diaphragm EMG can be recorded best using an esophageal catheter with multiple electrodes and has already proven its use as a powerful research tool (35). Today, the (processed) EMG signal can be obtained rather easily and continuously in ICU patients, which opens windows for using this tool in patient monitoring. The processed signal is from here referred to as the amplitude of the electrical activity of the diaphragm (EAdi). EAdi may be helpful in monitoring respiratory muscle loading, patient-ventilator synchrony and efficiency of breathing in critically ill patients.

#### Respiratory muscle loading

An important goal of mechanical ventilation is to unload the respiratory muscles. The ratio of actual EAdi to maximum EAdi is a measure of the patient’s effort to breath, where maximum EAdi can be defined as the peak activity observed during a 20 second inspiratory occlusion (27). A too low ratio suggests too high level of support, whereas a high ratio strongly suggests inadequate unloading of the respiratory muscles. In controlled mechanical ventilation, disuse of the diaphragm results in atrophy within a few days (11). High levels of pressure support may also suppress output from the respiratory centers by over-assistance resulting in respiratory muscle atrophy and contractile dysfunction (36, 37). In ARDS, extracorporeal membrane oxygenation (ECMO) improves oxygenation and carbon dioxide elimination. ECMO sweep gas flow can be adjusted to regulate carbon dioxide elimination. In patients recovering from ARDS, EAdi increases in response to decreasing ECMO sweep gas flow and vice versa (38). In selected cases monitoring of EAdi during ECMO allows titration of sweep gas flow to prevent extreme unloading of the diaphragm. Besides an increase in EAdi, high inspiratory loading and/or low inspiratory muscle capacity
Monitoring of the respiratory muscles in the critically ill

Figure 2. Screenshots from a mechanical ventilator in pressure support mode, tracings from top to bottom: airway pressure (Paw), airway flow, tidal volume, and diaphragm electrical activity (EAdi). (A) Over-assistance with pressure support ventilation leads to suppressions of diaphragm activity. (B) Diaphragm electrical activity reveals severe trigger delays (gray shaded area) and wasted efforts (indicated by arrows).
is also characterized by recruitment of the accessory inspiratory muscles. Data show that with decreasing levels of support the accessory muscle are increasingly activated (39). Although clinical implications of this observation are still unclear, it seems reasonable to assume that an increase in accessory muscle recruitment is the result of inadequate unloading. In the future, activity of accessory muscle may play a role in determining optimal ventilator settings.

**Patient-ventilator synchrony**

To date, visual inspections of flow and pressure waveforms are used to detect patient-ventilator asynchronies. However, it has been shown that the ability of ICU physicians, even experts, to do so is overall quite low and decreases at higher prevalence of asynchronies (40). Since EAdi is a direct measure of neural respiratory drive, it can be used to detect the onset and duration of neural inspiration and expiration. Consequently, monitoring EAdi could be considered the gold standard for detection of patient-ventilator asynchronies, including trigger delays, early and late cycling off, auto triggering, double triggering, and wasted efforts (Figure 2).

**Neuro-ventilatory and -mechanical efficiency**

Currently, new indices are evaluated for respiratory muscle function in ventilated patients. The ratio between tidal volume (Vt) and EAdi, represents neuro-ventilatory efficiency (NVE) of the diaphragm. An improved NVE indicates that a patient is able to generate the same Vt with lower levels of EAdi, whereas a higher EAdi suggests the opposite. NVE has been used to discriminate between extubation failure and success in patients weaning from mechanical ventilation (41). Evidently, NVE is sensitive to changes in diaphragm function (atrophy, fatigue and hyperinflation) as well as a patient’s load of breathing (airway compliance and resistance). Monitoring the ratio between Pdi and EAdi, represents neuro-mechanical efficiency (NME), excludes the influence of a patient’s load of breathing. A gradual decrease in NME over days indicates the development of diaphragm weakness, whereas an increase suggests recovery.

**Limitations**

Diaphragm electromyography is feasible in routine clinical care for monitoring respiratory muscle function. However, we still face several challenges. First, there is the need to identify the target level of EAdi or NVE that should be attained in the day-to-day management of mechanically ventilated patients. Second, it should be recognized that respiratory muscle activity is also suppressed by sedatives, in particular propofol and morphine-like analgetics (42). Third, insertion of an esophageal catheter...
carries a (low) risk for complications, and may be an uncomfortable procedure in non-sedated patients. Nevertheless, most critically ill patients require a naso-gastric tube for feeding, which are already commercially available with EMG electrodes (Maquet Critical Care, Solna, Sweden). Taken together, EAdi is a rational parameter to monitor respiratory muscle unloading and patient-ventilator synchrony from an early phase of critical illness. We expect that in the near future EAdi will become an important tool for respiratory muscle monitoring during mechanical ventilation and weaning (41, 43).

D. Phrenic nerve stimulation
Magnetic phrenic nerve stimulation allows non-voluntary evaluation of diaphragm strength, measured as the magnitude of twitch transdiaphragmatic pressure ($P_{di,tw}$) or twitch airway pressure ($P_{aw,tw}$) (44). The integrity of the phrenic nerve, in response to stimulation, can be tested by recording the compound muscle action potential (CMAP) of the diaphragm (45). This allows calculation of phrenic nerve conduction time and subsequent detection of phrenic nerve injury. Phrenic nerve stimulation has been extensively used in research settings. The technique, however, is not applicable for routine bed-side monitoring because of the fairly invasive nature, technical difficulties, and limitations concerning patients’ condition and tolerance. Therefore, this technique should be restricted for diagnostic purposes to a selection of patients (difficult weaning from mechanical ventilation) in centers that have sufficient experience with this technique.

E. Imaging
Radiography
Conventional radiography can be used for evaluating the position (chest x-ray) and motion (fluoroscopy) of the diaphragm. Elevation of the hemidiaphragm may be seen with hemidiaphragm paralysis. The clinical utility of this finding, however, is limited as hemidiaphragmatic elevation can occur in the absence of paralysis, such as with atelectasis, pneumonia and diaphragmatic eventration. Compared to chest x-ray, fluoroscopy provides more dynamic information and can be used to detect unilateral diaphragm paralysis. However, fluoroscopy is less helpful when the hemidiaphragm is weak but not completely paralyzed, and it can be particularly misleading in patients with bilateral diaphragmatic paralysis (8). Moreover, fluoroscopy is not suitable as a bedside monitoring tool.
Ultrasonography
Several recent studies have demonstrated the utility of ultrasonography for diaphragm muscle imaging. B-mode ultrasonography using a linear array transducer can be used to assess the diaphragm thickness in the zone of apposition (46, 47), an echogenic layer bordered by pleural and peritoneal membranes. Another promising method is the non-invasive evaluation of diaphragm motion with M-mode ultrasonography. With this mode, a phased array transducer is positioned subcostal or low intercostal between the midclavicular and mid-axillary lines. Procedure, reproducibility and normal values have been assessed recently in a large population of healthy subjects (9). In Figure 3, M-mode images are shown from a patient with normal diaphragm motion and diaphragm paralysis. Ultrasonography takes little time to perform, is noninvasive and can be performed at the bedside. However, diaphragm ultrasonography will only provide useful information during unassisted breathing and is therefore of limited relevance in the very early course of critical illness.

![Ultrasonographic M-mode images from the right hemidiaphragm.](image)

**Figure 3.** Ultrasonographic M-mode images from the right hemidiaphragm. (A) Normal diaphragm motion, the diaphragm moves 1.8 cm cephalad during inspiration. (B) Caudal movement of the diaphragm in a patient with diaphragm paralysis.

More complex imaging techniques, such as magnetic resonance imaging (MRI) and computed tomography (CT) have been used to evaluate diaphragm function (48, 49), but are not suitable for monitoring as these techniques are cumbersome in mechanically ventilated patients and should only be used for specific diagnostic purposes.
F. Circulatory biomarkers

Studies in critically ill patients and ventilated animals revealed that respiratory muscle weakness is associated with muscle fiber damage and loss of contractile proteins (2, 11, 14, 16, 17). Considering the important role of plasma markers in the diagnosis of myocardial damage and rhabdomyolysis, it could be reasoned that the detection of muscle specific proteins in plasma would be a valuable tool to monitor damage of the respiratory muscles. Although potential candidates include classical markers such as creatine kinase and myoglobin, the clinical use of these markers is probably hampered by their large range in healthy individuals and non-specificity for structural damage (50). Recent experimental findings indicate that measuring serum levels of skeletal muscle troponin I is more sensitive to detect structural injury of respiratory muscles (51). However, the relation between plasma troponin I levels and functional measures in critically ill patients has not yet been investigated. Of note, respiratory muscle weakness in the critically ill is often part of generalized muscle weakness. In those cases, evaluation of circulatory biomarkers of skeletal muscle damage may not specifically reflect the functioning of respiratory muscles, but could rather represent overall skeletal muscle function.

How could respiratory muscle monitoring affect clinical care?

Given the detrimental effects of critical illness, monitoring respiratory muscle function is a reasonable approach, even in the absence of evidence based therapeutic implications. If monitoring indicates rapid loss of respiratory muscle function then factors that negatively affect muscle function should be restricted as much as possible, including disuse by over-assistance, patient-ventilator asynchrony and drugs associated with side effects on muscle function. In fact, this is to a certain extent in analogy with monitoring renal function in critically ill patients. There are no direct therapeutic implications in monitoring renal function, but clinicians will avoid certain nephrotoxic drugs when renal function is impaired.

Monitoring in the early phase

In the early phase of critical illness, monitoring of the respiratory muscles should focus on minimizing the negative effects of mechanical ventilation. During this phase, clinicians should avoid too high (‘over-assist’) as well as too low levels of pressure support. Although the width of this therapeutic margin is unknown, monitoring EAdi might prove helpful to detect suppressed output from the respiratory centers by
over-assistance as well as excessive respiratory drive during inadequate unloading. A considerable group of mechanically ventilated patients (25%) exhibit severe patient-ventilator asynchrony (52). Such a high incidence is associated with a prolonged duration of mechanical ventilation (52, 53). Hence, early detection of severe asynchrony by monitoring EAdi can be used to adjust ventilator settings to improve synchrony. For example, neutrally adjusted ventilatory assist (NAVA) has been shown to reduce patient-ventilator asynchrony (54). In the early phase of ARDS the use of neuromuscular blocking agents (NMBA) has been shown to improve outcome (55). Muscle paralysis prevents patient-ventilator asynchrony, thereby reducing the risk of baro- and volutrauma, providing an acceptable explanation for the beneficial effects observed in this latter study (56). If true, monitoring EAdi to detect patient-ventilator asynchrony, during brief interruption of NMBA, may help the clinician to decide when to stop further NMBA administration. Also, we use EAdi to titrate NMBA in selected ARDS patients. A bolus is administered when EAdi is observed, thereby reducing the risk of overdose.

Monitoring during (difficult) weaning
During weaning from mechanical ventilation, short periods of unassisted breathing allow closer monitoring of respiratory muscle function using ultrasonography and respiratory pressure measurements. Using M-mode ultrasonography, diaphragm dysfunction was found in 29% of critically ill patients without history of diaphragm dysfunction (57). In a non-ICU population, it was shown that sequential measurements of diaphragm thickness using B-mode ultrasonography is not only useful in making the initial diagnosis of diaphragm weakness, but for determining subsequent recovery as well (58). Knowing whether or not the respiratory muscles are impaired is of major clinical importance. It allows development of an effective treatment plan (including mobilization and physiotherapy) and proper prognostic advice to the patient and relatives.

Therapeutic strategies
Contrary to another vital muscle, the heart, there are currently limited strategies available to improve respiratory muscle function. However, data from recent pre-clinical studies offer hope for the near future (59-61). Recently, we have shown that the calcium sensitizer levosimendan improves neuro-mechanical efficiency and contractile function of the human diaphragm in vivo. Our findings suggest a new therapeutic approach to improve respiratory muscle function in patients with respiratory failure (59). In mechanically ventilated rats, the antioxidant N-acetylcysteine prevented ventilation induced diaphragmatic oxidative stress and proteolysis and
abolished ventilation-induced diaphragmatic contractile dysfunction (60). Like any skeletal muscle, the diaphragm is responsive to training. In a randomized controlled trial, inspiratory muscle training (IMT) has been shown to improve inspiratory muscle strength and weaning outcome in critically ill patients (61). IMT should therefore be considered in difficult to wean patients with proven inspiratory muscle weakness.

Further studies are needed to establish the effects of these new pharmacological interventions and to develop specific training protocols for critically ill patients. Adequate monitoring of respiratory muscle function is indispensible to establish the effects of these interventions.

Summary and future directions

A large body of evidence shows the detrimental effects of critical illness on respiratory muscle structure and function, which is associated with prolonged weaning from mechanical ventilation. While some factors related to impaired muscle function in the critically ill are unpreventable (e.g. sepsis), awareness of developing weakness may alter treatment (e.g. ventilator settings, training, drug prescription).

As outlined, different tools are available to assess respiratory muscle function. Some of these tools have limited value (chest x-ray, fluoroscopy) or are not suitable for routine clinical care monitoring (MRI, CT, phrenic nerve stimulation). However, more promising tools that can be used today or in the near future for monitoring of respiratory muscle function are: (1) ultrasonography to evaluate diaphragm movement and thickness; (2) measurement of mouth and/or transdiaphragmatic pressure to monitor respiratory muscle strength; and (3) electromyography of the diaphragm to monitor respiratory muscle unloading and patient-ventilator asynchrony. Circulatory biomarkers for respiratory muscle injury are much needed and hopefully will appear in the next few years.

There is scarce literature that directly demonstrates improved outcome with close monitoring (and action) of the respiratory muscles. However, over the last years circumstantial evidence suggests that respiratory muscle monitoring can affect clinical care in the ICU.

Today, we are only at the beginning of routinely monitoring respiratory muscle function. However, practical issues and the absence of sound scientific data for clinical benefit should in our opinion not discourage clinicians for having a closer look at respiratory muscle function in critically ill patients. In modern ICUs, monitoring the respiratory muscles should be as much part of the routine as monitoring any other organ function.
References


Monitoring of the respiratory muscles in the critically ill


Chapter 3

The differential diagnosis for failure to wean from mechanical ventilation

Jonne Doorduin, Johannes G. van der Hoeven, Leo M.A. Heunks

*Curr Opin Anaesthesiol. 2016 Apr;29(2):150-7*
Abstract

Purpose of review: In this review, we discuss the causes for a failed weaning trial and specific diagnostic tests that could be conducted to identify the cause for weaning failure. We briefly highlight treatment strategies that may enhance the chance of weaning success.

Recent findings: Impaired respiratory mechanics, respiratory muscle dysfunction, cardiac dysfunction, cognitive dysfunction and metabolic disorders are recognized causes for weaning failure. In addition, iatrogenic factors may be at play. Most studies have focused on respiratory muscle dysfunction and cardiac dysfunction. Recent studies demonstrate that both ultrasound and electromyography are valuable tools to evaluate respiratory muscle function in ventilated patients. Sophisticated ultrasound techniques and biomarkers such as BNP, are valuable tools to identify cardiac dysfunction as a cause for weaning failure. Once a cause for weaning failure has been identified specific treatment should be instituted. Concerning treatment, both strength training and endurance training should be considered for patients with respiratory muscle weakness. Inotropes and vasodilators should be considered in case of heart failure.

Summary: Understanding the complex pathophysiology of weaning failure in combination with a systematic diagnostic approach allows identification of the primary cause of weaning failure. This will help the clinician to choose a specific treatment strategy and therefore may fasten liberation from mechanical ventilation.
The differential diagnosis for failure to wean

Introduction

Weaning from mechanical ventilation covers the entire process of liberating a patient from the ventilator and the endotracheal tube (1). Weaning failure is usually defined as an unsuccessful spontaneous breathing trial (SBT) or need for ventilator support (including noninvasive ventilation) within 48 hours after extubation (1). In patients that require more than 7 days of weaning after a first failed spontaneous breathing trial, mortality is significantly increased (13% versus 7% of patients that need shorter weaning time (2)). The pathophysiology of a failed weaning trial may be complex, but it is in our opinion important to understand the reason for a failed weaning trial. Identification of the cause of weaning failure will help the clinician to choose a rationale treatment strategy that improves the chances of success for the next weaning trial. The aim of this paper is summarize the differential diagnosis of a failed weaning trial and discuss recent papers that help the clinician to identify the cause for weaning failure. Finally, we briefly discuss new treatment strategies that may fasten liberation from mechanical ventilation.

Causes of weaning failure

The transition from positive inspiratory pressure during mechanical ventilation to negative airway pressure during spontaneous breathing challenges the patients physiological reserve. When an imbalance develops between the patients’ ventilatory needs and capacity, weaning fails. Below we will discuss the causes of weaning failure and the techniques that are most helpful in the diagnostic work-up.

Impaired respiratory mechanics

The load imposed upon the respiratory muscles is determined by the resistance and compliance of the respiratory system and the presence of PEEPi. An increase in load is accompanied by an increase in work of breathing and as such may lead to weaning failure. Indeed airway resistance is higher in patients failing a weaning trial (3, 4). In the upper airways, resistance may be increased by the artificial airway and/or tracheal injury, including stenosis, tracheomalacia, and granulation tissue. In tracheostomized patients with weaning failure referred to a dedicated weaning center, endoscopy revealed significant tracheal stenosis (>50% of tracheal lumen) in 5% (14/288) of the patients (5). Ten of these 14 patients could be successfully decannulated after removal of granulation tissue.

The presence of elevated small airway resistance is obvious in patients with COPD or asthma, but in acute respiratory distress syndrome (ARDS) airway resistance may be increased as well due to edema of the bronchial wall. As a result of increased
airway resistance, expiratory flow is limited and consequently intrinsic positive
dered-expiratory pressure (PEEPi) may develop. PEEPi elevates work of breathing
via hyperinflation and by acting as a threshold for generation of inspiratory flow.
Under dynamic conditions, PEEPi can be determined only by using an esophageal
balloon. Although these balloons are widely available, positioning of the catheter
and interpretation of the signal is subject to pitfalls (6).

The compliance of the respiratory system is determined by the elastic properties
of both the lungs and the chest wall. In a population of ARDS patients, compliance
was found to be lower in the failure group compared to successful weaning (4).
Lung compliance may be reduced due to alveolar filling (edema or pus), atelectasis,
interstitial lung disease, pulmonary fibrosis, and hyperinflation. Pleural effusion,
edema, obesity and elevated abdominal pressure decrease chest wall compliance.
Measurement of esophageal pressure (Pes) is required to differentiate lung compliance
from chest wall compliance. However, in clinical practice when total respiratory
compliance is decreased clinical and / or radiological work up will generally identify
the cause of reduced compliance.

**Respiratory muscle dysfunction**

Critical illness has profound effects on respiratory muscle structure and function
(7). Rapid development of diaphragm weakness, indicated by approximately a third
reduction in diaphragm force, was found in the first 5-6 days of invasive mechanical
ventilation in a small group of critically ill patients (8). Diaphragm muscle fiber cross-
sectional area is reduced by ±25% in critically ill patients ventilated for ±7 days
compared to patients referred for elective surgery (9). In addition, in-vitro force
generation of these fibers was severely reduced compared to controls. Goligher and
colleagues (10) found that in 44% of the ventilated ICU patients diaphragm thickness,
as assessed by ultrasound, decreased by more than 10% in the first week on the
ventilator. The reduction in diaphragm thickness was associated with weakness (10).

Clinical evaluation of respiratory muscle function in ventilated critically ill patients
is a challenge. Previously, we discussed the available techniques in detail (11, 12).
Here, we will briefly discuss clinically relevant and feasible techniques. Maximal
inspiratory pressure (MIP) and maximal expiratory pressure (MEP) are tests of global
respiratory muscle strength and can be measured using a handheld device connected
to the artificial airway. A MIP above 30 cmH₂O is associated with a shorter time
to successful extubation (13). Simultaneous recording of Pes and gastric pressure
(Pga) using dedicated balloons allows calculation of transdiaphragmatic pressure
(Pdi = Pga - Pes), a specific measure of diaphragm contractility. The latter is useful
for close monitoring and evaluation of diaphragm function in difficult-to-wean
The differential diagnosis for failure to wean patients (Table 1). However, as mentioned before, acquisition and interpretation of Pes, Pga and their derived measures, such as work of breathing, requires expertise.

Diaphragm ultrasonography is a practical and non-invasive tool for assessment of diaphragm thickness, thickening fraction and displacement (Figure 1). Using M-mode ultrasonography, diaphragmatic dysfunction (vertical excursion < 1 cm or paradoxic movements) was found in 24 of 82 patients who met criteria for an SBT (14).

Using M-mode ultrasonography, diaphragmatic dysfunction (vertical excursion < 1 cm or paradoxic movements) was found in 24 of 82 patients who met criteria for an SBT (14). These patients showed frequent early and delayed weaning failures. DiNino and colleagues found in 63 mechanically ventilated patients that diaphragm thickening fraction above 30% predicts extubation success with a sensitivity and specificity of 88% and 71%, respectively (15). Very recently, it was proposed that thickening fraction of the diaphragm as assessed with ultrasound could be used to detect dysfunction (10). Future fields of ultrasonography application may be detection of patient-ventilator asynchrony (16), and assessment of respiratory workload (17, 18).

Diaphragm electromyography (EMG) reflects neural respiratory drive to the diaphragm. The (processed) EMG signal can be obtained continuously using commercially available multi-electrode esophageal catheters required for neurally adjusted ventilatory assist (NAVA) ventilator mode (Maquet, Solna, Sweden). The ratio between diaphragm EMG and tidal volume is called the neuro-ventilatory efficiency (NVE). NVE has been shown to identify weaning failure in multiple studies (19-22). Evidently, NVE is sensitive to changes in diaphragm function as well as a patient’s load of breathing. Diaphragm EMG may also be used to monitor respiratory muscle unloading (23) patient-ventilator interaction (24) and the effect of residual sedation on respiratory drive (25). In a preliminary observation, it was reported that diaphragm EMG can be used to detect development of fatigue after extubation (26).

Cardiac dysfunction
The lungs and heart are functionally and anatomically coupled and therefore transition of assisted breathing to unassisted breathing has profound cardiovascular effects that may induce weaning failure. First, during positive pressure ventilation the intra-thoracic pressure (ITP) increases during inspiration, whereas during unassisted breathing ITP decreases during inspiration as a result of activation of the inspiratory muscles. The decrease in ITP during a weaning trial will enhance venous return by reducing right atrial pressure, end diastolic RV volume and such in patients with normal cardiac function increase RV and LV output, but may result in heart failure in patients with compromised cardiac function. Second, increased
sympathetic tone (emotional stress, hypercapnia, hypoxemia) may further increase LV afterload. Finally, the transition to unassisted breathing puts an elevated load on the respiratory muscles, which increases oxygen consumption and such is a stress to the heart. The final effects of unassisted breathing on the cardiovascular system are much more complex. We refer to recent papers for review (27, 28). Table 2 shows the cardiovascular effects of a successful prolonged spontaneous breathing trail in a typical ICU patient.

As an alternative to the pulmonary artery catheter, less invasive techniques are currently used to identify cardiac failure during a weaning trial. Transthoracic echocardiography during an SBT can identify both systolic and diastolic dysfunction (29). With tissue Doppler imaging, myocardial relaxation can be quantified by measuring the early diastolic mitral annulus velocity (Ea). In combination with transmitral early diastolic filling velocity (E), measured by conventional pulsed wave

---

**Figure 1.** Normal diaphragm ultrasonography. From left-to-right, top-to-bottom: B-mode diaphragm thickness during expiration, B-mode diaphragm thickness during inspiration, M-mode diaphragm displacement during normal inspiration, measurements obtained from images. Abbreviations: Dia = diaphragm; exp = expiration; ICS = intercostals muscles; insp = inspiration; SC = subcutis.

ultrasonography measurements:
- thickness exp. = 1.6 mm
- thickness insp. = 2.4 mm
- thickening fraction = $(2.4 - 1.6) / 1.6 = 50\%$
- displacement = 2.3 cm
Doppler, the resulting E/Ea ratio closely correlates with left ventricular filling pressure (30). Moschietto and colleagues found in a non-selected population of 68 patients, performing an SBT, that the E/Ea ratio before and during the SBT was higher in the failure group than in the successful group (29). In this study, an E/Ea ratio during the SBT of 14.5 predicted weaning failure with a sensitivity of 75% and a specificity of 95.8%. It should be recognized that performing ultrasound at the time of weaning failure might be a quite challenge.

B-type natriuretic peptide (BNP) and N-terminal (NT)-proBNP are cardiac biomarkers secreted by ventricular cardiomyocytes in response to increased ventricular wall stress. Increases in BNP and NT-proBNP levels during the SBT are consistent with weaning failure of cardiac origin, in which BNP performs better than NT-proBNP (31, 32). An increase in BNP levels by more than 12% allowed diagnosing weaning failure from cardiac origin with a sensitivity of 76% and a specificity of 78%, with the pulmonary artery catheter as a reference method (32). Mekontso Dessap and colleagues showed that a BNP-guided fluid management strategy was associated with a shorter duration of mechanical ventilation, especially in patients with left ventricular systolic dysfunction (33). In conclusion, changes in BNP levels are a reliable and feasible alternative to the pulmonary artery catheter for diagnosing weaning-induced pulmonary edema. A passive leg raising before the SBT has also been suggested to identify weaning failure patients related to cardiac dysfunction (34). Recently, Dres and colleagues developed an algorithm for management of weaning failure from cardiac origin (27).

Table 1. Respiratory effects of a spontaneous breathing trial.

<table>
<thead>
<tr>
<th></th>
<th>Resp rate (l/min)</th>
<th>Vt (ml)</th>
<th>Edi (µV)</th>
<th>∆Pes (cmH₂O)</th>
<th>∆Pga (cmH₂O)</th>
<th>∆Pdi (cmH₂O)</th>
<th>PEEPi (cmH₂O)</th>
<th>WOB (J/L)</th>
<th>PaO₂ (mmHg)</th>
<th>PaCO₂ (mmHg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PS *</td>
<td>34</td>
<td>211</td>
<td>20</td>
<td>12</td>
<td>1</td>
<td>13</td>
<td>5</td>
<td>1.0</td>
<td>75</td>
<td>56</td>
</tr>
<tr>
<td>5 min</td>
<td>36</td>
<td>246</td>
<td>48</td>
<td>25</td>
<td>2</td>
<td>26</td>
<td>10</td>
<td>2.1</td>
<td>61</td>
<td>56</td>
</tr>
<tr>
<td>10 min</td>
<td>37</td>
<td>260</td>
<td>43</td>
<td>24</td>
<td>2</td>
<td>26</td>
<td>9</td>
<td>2.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>60 min</td>
<td>23</td>
<td>352</td>
<td>52</td>
<td>30</td>
<td>2</td>
<td>32</td>
<td>10</td>
<td>2.6</td>
<td>70</td>
<td>57</td>
</tr>
</tbody>
</table>

Successful spontaneous breathing trial in a COPD patient despite an increase in PEEPi and WOB. Abbreviations: Edi = electrical activity of the diaphragm; PaCO₂ = arterial carbon dioxide partial pressure; PaO₂ = arterial oxygen partial pressure; Pdi = transdiaphragmatic pressure; PEEPi = intrinisic positive end-expiratory pressure; Pes = esophageal pressure; Pga = gastric pressure; Resp rate = respiratory rate; Vt = tidal volume; WOB = work of breathing. * PS = 5 cmH₂O, PEEP = 10 cmH₂O
Table 2. Cardiovascular effects of a spontaneous breathing trial.

<table>
<thead>
<tr>
<th></th>
<th>SvO2 (%)</th>
<th>PCWP (mmHg)</th>
<th>PAP (mmHg)</th>
<th>ABP (mmHg)</th>
<th>CVP (mmHg)</th>
<th>HR (/min)</th>
<th>SpO2 (%)</th>
<th>PaO2 (mmHg)</th>
<th>CO (L/min)</th>
<th>BNP (pg/mL)</th>
<th>Lactate (mmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PS *</td>
<td>64</td>
<td>22</td>
<td>27/45 (35)</td>
<td>59/124 (79)</td>
<td>15</td>
<td>63</td>
<td>94</td>
<td>75</td>
<td>5.3</td>
<td>1358</td>
<td>1.0</td>
</tr>
<tr>
<td>5 min</td>
<td>62</td>
<td>23</td>
<td>27/40 (33)</td>
<td>67/158 (97)</td>
<td>11</td>
<td>68</td>
<td>92</td>
<td>61</td>
<td>6.4</td>
<td></td>
<td></td>
</tr>
<tr>
<td>60 min</td>
<td>65</td>
<td>25</td>
<td>25/42 (34)</td>
<td>70/163 (100)</td>
<td>10</td>
<td>70</td>
<td>94</td>
<td>70</td>
<td>6.3</td>
<td>1450</td>
<td>1.1</td>
</tr>
</tbody>
</table>

Successful spontaneous breathing trial in a COPD patient with a normal cardiovascular response. Abbreviations: ABP = arterial blood pressure; BNP = B-type natriuretic peptide; CO = cardiac output; CVP = central venous pressure; HR = heart rate; PaO2 = arterial oxygen partial pressure; PAP = pulmonary artery pressure; PCWP = pulmonary capillary wedge pressure; SvO2 = mixed venous oxygen saturation. * PS = 5 cmH2O, PEEP = 10 cmH2O

Cognitive dysfunction
A serious and frequent psycho-organic disorder in critically ill patients is delirium. Critically ill patients with a delirium have a 7 times higher risk of prolonged mechanical ventilation (35). Moreover, it was found that delirium was associated with more respiratory and neurologic complications and a reduced probability of successful extubation (36). The CAM-ICU is a validated tool to detect delirium (37). Besides delirium other psychological factors may play a role in weaning failure. It should be recognized that the inability to communicate physical and emotional needs, and ventilator-dependence cause mental stress and contribute to weaning failure (38). Depressive disorders are associated with weaning failure as well (39). Consultation of a psychiatrist is reasonable when a psychological disorder is suspected.

Endocrine and metabolic disorders
The role of endocrine dysfunction in weaning failure has never been systematically evaluated, but is most likely of limited importance. Hypothyroidism and adrenal insufficiency should be excluded in case of clinical suspicion, as treatment options are available. Malnutrition frequently occurs in critically ill patients and is associated with higher mortality (40), but also with reduced muscle mass and as such contributes to difficult weaning, as discussed above.

Iatrogenic factors
The role of the clinician in weaning is crucial, in particular the assessment whether the patient still requires ventilator support. In patients with unplanned extubation only
44% required reintubation (41). When unplanned extubation occurred during the weaning phase, only 30% required reintubation. Patients with successful unplanned extubation during weaning phase spent less time on weaning the ventilator (41). In a large clinical trial comparing weaning methods performed in a long-term weaning facility (42), 32% of the patients passed the initial screening weaning trial. This indicates that these patients were already weaned upon arrival in this weaning facility, although this was unrecognized by the referring ICU team. These studies stress the importance to screen patients for readiness for weaning and extubation. Appropriate tests for weaning readiness have recently been reviewed elsewhere (43). Under certain conditions there may be a role for automated weaning (44).

Asynchrony between the patient and ventilator is associated with increased duration of weaning (45) and even mortality (46). Although it is reasonable to aim for optimal patient-ventilator interaction, no studies have been performed to study the effect on weaning outcome.

Therapeutic consequences
Once a specific reason for weaning failure has been identified, the clinician should develop a strategy that increases the chances of successful weaning.

Respiratory mechanics
In patients with elevated airway resistance bronchodilators should be administered. Under certain conditions (malpositioning, granulation tissue) the tracheostomy can enhance airway resistance (47). Specifically adapted tubes may be required. In patients with impaired respiratory compliance it is often obvious how mechanics can be improved, such as evacuation of pleural fluid or ascites, diuretics to achieve negative fluid balance in case of chest-wall or pulmonary edema (48).

Respiratory muscle dysfunction
Disuse is a prominent risk factor for the development of respiratory muscle weakness as pointed out above. Therefore, it is reasonable to limit the duration of respiratory muscle inactivity, by using assisted modes for ventilation. Future studies should determine the optimum time point for initiation of assisted ventilation and the level of unloading of the inspiratory muscles.

Respiratory muscle strength training seems as a reasonable intervention in weak patients. Most experience for respiratory muscle training has been obtained in patients with COPD. For instance in a randomized controlled trial (N=33; not on the ventilator) inspiratory muscle strength training improved respiratory muscle strength, 6-minute walking distance and reduces dyspnea sensation, compared to
sham training (49). Very few studies have been performed in ventilated patients. The best evidence up to now comes from Martin and colleagues, demonstrating that inspiratory muscle training in long-term ventilated patients improves muscle strength and the chances of successful weaning (50). Although many questions remain concerning optimal timing and protocol (51), initiation of inspiratory muscle training appears reasonable in weak ventilated ICU patients difficult to wean from the ventilator.

In contrast to cardiac muscle dysfunction, no drug is approved to optimize respiratory muscle function. However, some small pilot studies demonstrate that respiratory muscle contractility can be enhanced pharmacologically (52). Levosimendan is a relatively novel cardiac inotrope that improves calcium sensitivity of the contractile proteins. In healthy subjects levosimendan improves contractile efficiency of the respiratory muscles (53). A clinical trial in difficult to wean patients is currently conducted (NCT01721434). Anabolic hormones that directly enhance contractile protein synthesis are another potentially interesting strategy to enhance respiratory muscle function. In a randomized controlled trial Schols demonstrated that nandrolone together with high caloric feeding enhances respiratory muscle strength in patients with COPD participating in a rehabilitation program (54). The effect of anabolic steroids on peripheral or respiratory muscle strength in difficult to wean ICU patients has not been investigated and the routine use is not recommended today.

Cardiac dysfunction
Routsi evaluated the effects of nitroglycerin infusion on hemodynamics and weaning outcome in difficult to wean patients with COPD and arterial hypertension (55). In these patients, nitroglycerin had beneficial hemodynamic effects during a spontaneous breathing trial, including reduction in pulmonary capillary wedge pressure, right ventricular stroke work index and improved venous oxygen saturation. Moreover, whereas all patients (12) failed a weaning trial under control conditions, 92% successfully completed the SBT under nitroglycerin infusion. In patients with overt heart failure inotropes could be considered, despite the absence of strong evidence. Levosimendan has been shown to improve cardiac output and reduce pulmonary capillary wedge pressure and clinical outcome in patients with acute heart failure (56, 57). The effects of levosimendan on weaning outcome was evaluated in a small pilot study, including 12 patients with systolic heart failure (left ventricular ejection fraction 28% ± 5%) (58). After levosimendan infusion 7 of the 12 patients were weaned from mechanical ventilation within 51 hours of treatment. Although there is a strong rationale for inotropes in patients with heart failure that are difficult
to wean from the ventilator, clinical studies should confirm the favorable effect on outcome.

**Cognitive dysfunction**
Reducing the level of sedation has been shown to reduce duration of mechanical ventilation (59). More recently, in a propensity score analysis Lonardo demonstrated that the use of propofol instead of benzodiazepines reduced duration of mechanical ventilation and mortality in a heterogeneous group of patients (60). Among other factors, it should be recognized that benzodiazepines are associated with the development of delirium (61). In addition, benzodiazepines adversely affect sleep architecture as assessed by polysomnography in weaning patients (62). In patients with confirmed depressive disorder that affect weaning, treatment with psychostimulants may be considered (63).

**Endocrine and metabolic disorders**
The role of thyroid hormone replacement therapy in difficult to wean patients has not been systemically evaluated. However, suppletion of thyroid hormone in patients with proven hypothyroidism is reasonable (64). Huang and colleagues have demonstrated that in patients with confirmed adrenal insufficiency treatment with hydrocortisone fastens weaning from the ventilator (65). No specific nutritional therapy has been shown to improve weaning outcome.

**Conclusion**
The pathophysiology of weaning failure is complex and requires a systematic differential diagnostic approach to identify the primary cause for weaning failure. Several tools and techniques are available to discriminate between these causes. Identification of the cause of weaning failure allows specific treatment and thereby may fasten liberation from mechanical ventilation.
References


The differential diagnosis for failure to wean


The differential diagnosis for failure to wean
Chapter 4

Inspiratory and expiratory muscle effort during successful and failed weaning from mechanical ventilation

Jonne Doorduin, Lisanne H. Roesthuis, Diana Jansen, Johannes G. van der Hoeven, Hieronymus W.H. van Hees, Leo M.A. Heunks

Submitted for publication
Abstract

**Background:** Respiratory muscle dysfunction is an important cause for difficult weaning from mechanical ventilation. Today, several studies have evaluated inspiratory muscle function in weaning patients. Although the expiratory muscles play an important role in breathing, in particular in patients with inspiratory muscle weakness or high breathing effort, the role of the expiratory muscles during weaning in ICU patients has not been studied in detail. The current physiological study aims to compare the recruitment of the inspiratory muscles and expiratory muscles during weaning.

**Methods:** Twenty-one adult patients receiving pressure support ventilation underwent a spontaneous breathing trial (T-tube with supplemental oxygen) of maximal 1 hour. During the trial flow, gastric pressure, esophageal pressure and diaphragm electrical activity were recorded continuously. Transdiaphragmatic pressure, neuromechanical efficiency of the diaphragm, pressure-time product of the inspiratory muscles and expiratory muscles were calculated.

**Results:** Ten patients failed the spontaneous breathing trial. In the weaning failure group diaphragm electrical activity was higher (71 %; \(p<0.05\)) and neuro-mechanical efficiency of the diaphragm was lower (41 %; \(p<0.05\)). In the failure group the contribution of the expiratory muscles to total respiratory muscle effort increased up to 26±4 % during spontaneous breathing compared to 10±3 % in the weaning success group (\(p<0.05\)).

**Conclusions:** The expiratory muscles significantly contribute to respiratory muscle effort in critically ill patients who fail a trial of spontaneous breathing. In addition, our findings confirm that impaired pressure-generating capacity of the diaphragm, regardless of its origin, plays a central role in failure to wean from mechanical ventilation.
Introduction

Prolonged weaning from mechanical ventilation is associated with increased morbidity and mortality (1-3), and therefore, successful extubation is a crucial step for critically ill patients. However, approximately 6-15% of mechanically ventilated patients experience prolonged weaning (1-4). A major determinant of weaning failure is respiratory muscle dysfunction (5, 6). The respiratory muscles, in particular the diaphragm, are profoundly affected by critical illness and mechanical ventilation (7-13). Only a few days of controlled mechanical ventilation is associated with a decline in diaphragmatic pressure generating capacity (10). In particular with increased respiratory load, diaphragm weakness may result in failure to wean from mechanical ventilation (14).

Detailed analysis of diaphragm function by recording diaphragm electromyography (EMGdi) and transdiaphragmatic pressure (Pdi) during a spontaneous breathing trial (SBT) may help to understand the role of diaphragm dysfunction in patients who fail to wean from mechanical ventilation. These measurements allow calculation of the so-called neuromechanical efficiency (NME) of the diaphragm, i.e. the ability of the diaphragm to convert neural input into mechanical output (15). Recordings of esophageal pressure (Pes) also allows in depth analysis of respiratory mechanics and muscle effort (16, 17). Gastric pressure (Pga) is used to calculate Pdi and also the recruitment of the abdominal muscles during expiration. Expiratory muscle recruitment may be a mechanism for offsetting the effects of increased load on a weak diaphragm and to decrease end-expiratory lung volume to limit hyperinflation (18). In patients with chronic obstructive pulmonary disease (COPD) expiratory muscle recruitment has been demonstrated during weaning failure (19, 20). However, it is unknown to what extent expiratory muscle recruitment contributes to total respiratory muscle effort in weaning patients.

Accordingly, the aim of the current physiological study was to compare the recruitment of the inspiratory muscles and expiratory muscles in weaning success and weaning failure. Additionally, we performed detailed analysis of respiratory mechanics in these patients.

Methods

Study design and population
This cross-sectional physiological study was conducted in the intensive care unit (ICU) of the Radboud university medical center Nijmegen, the Netherlands. Twenty-
one adult patients receiving invasive mechanical ventilation for at least 3 days and considered ready for a spontaneous breathing trial were recruited. The decision to extubate or resume mechanical ventilation was made solely by the clinical team, who were blinded to the study data. Exclusion criteria were a medical history of neuromuscular disorders, upper airway or esophageal pathology (e.g. recent surgery, esophageal varices, diaphragmatic hernia) and recent (< 1 month) nasal bleeding.

The protocol was approved by the local ethics review committee (approval number: 2010-058) and conducted in accordance with the Declaration of Helsinki and its later amendments. Written informed consent was obtained from patients who did not have a multi-electrode esophageal catheter with two balloons in situ prior to the start of the study.

**Study protocol**

All patients were ventilated with the Servo-i ventilator (Maquet Critical Care, Sölna, Sweden). If not already in situ, a multi-electrode esophageal catheter with two balloons (NeuroVent Research Inc, Toronto, Canada) was inserted nasally. Catheter characteristics and positioning techniques have been described previously (15, 21, 22). Next, 10 minutes of pressure support ventilation (PSV) was recorded with an inspiratory support level of 8 cmH$_2$O and a positive end-expiratory pressure (PEEP) level of 5 cmH$_2$O. Subsequently, a spontaneous breathing trial, using a T-tube with supplemental oxygen, was performed for up to 60 minutes, according to our clinical protocol. The criteria for weaning failure were tachypnea (> 35 breaths / min), low arterial oxygen saturation (< 90%), tachycardia (> 140 beats / min), hypertension (systole > 180 mmHg), hypotension (systole < 90 mmHg), agitation, diaphoresis, and anxiety during the SBT. Patients were extubated after a successful SBT. Weaning failure was defined as a unsuccessful SBT, or reintubation within 48 hours after extubation. Arterial blood samples were collected before disconnection from the ventilator and at the end of the SBT before reinstitution of ventilator support.

**Data acquisition**

Flow was measured with a Fleisch pneumotachograph (Hans Rudolph, Kansas City, MO, USA) placed at the endotracheal tube or cannula. The pneumotachograph was connected to a differential pressure transducer (range ± 50 kPa, Freescale, Tempe, AR). Esophageal pressure (Pes) and gastric pressure (Pga) were obtained using two thin-walled balloons filled with air attached on the esophageal catheter. The balloons were connected to two differential pressure transducers (range ± 50 kPa, Freescale, Tempe, AR). Pressure signals from the transducers were digitized (Porti 16, 22 bits,
1.4 µV/least significant bit, TMSi; The Netherlands) at a sampling frequency of 100 Hz. Pdi was calculated as Pes subtracted from Pga.

EMGdi signals recorded from the electrodes on the esophageal catheter were amplified and digitized (Porti 16, 22 bits, 71.5 nV/least significant bit, TMSi; the Netherlands) at a sampling frequency of 2 kHz. EMGdi signal processing from the multiple electrodes was performed according to the method of Sinderby and colleagues (23-25).

Flow, Pes, Pga and EMGdi were acquired synchronously using dedicated software (NeuroVent Research Inc, Toronto, Canada) and stored on a hard disk for offline analysis in a software routine developed for Matlab (R2014b, The Mathworks, Natick, MA).

**Physiological measurements**

Inspiratory time (Ti), expiratory time (Te), total respiratory cycle time (Ttot) and respiratory rate were derived from the flow signal. Tidal volume (Vt) was calculated as the integral of inspiratory flow.

\[ \Delta \text{Pes} \] was calculated as the difference between the start of decrease in Pes and the negative peak value of Pes during inspiration. \[ \Delta \text{Pga} \] was calculated as the difference between the start of increase in Pga and the positive peak value of Pga during inspiration. \[ \Delta \text{Pdi} \] was calculated as the difference between the start of increase in Pdi and the positive peak value of Pdi during inspiration. \[ \Delta \text{Pes} \] and \[ \Delta \text{Pga} \] were corrected for the rise in expiratory gastric pressure (\[ \Delta \text{Pga}_{\text{exp}} \]) in the preceding breath, as described previously (20). \[ \Delta \text{Pga}_{\text{exp}} \] was calculated as the rise in gastric pressure from end-inspiration to the start of decrease in Pes (i.e. start of inspiration). PEEPi was calculated as the rise in Pdi until start of inspiratory flow and thus corrected for a drop in gastric pressure at start of inspiration. The Gilbert index, a measure of relative contribution of the diaphragm to inspiratory pressure, was calculated as \[ \Delta \text{Pga} / \Delta \text{Pdi} \]. Dynamic lung compliance (\( C_{\text{dyn}} \)) and inspiratory resistance of the lung and airways (Rinsp) were calculated as described previously (26).

\[ \Delta \text{EMGdi} \], an estimation of neural respiratory drive, was calculated as the peak root-mean-square of the EMGdi during inspiration. Center frequency of the EMGdi (CFdi), a measure of contractile fatigue of the diaphragm (27), was calculated using spectral analysis of the EMGdi. A detailed description of calculation of EMGdi and CFdi can be found in the work by Sinderby and colleagues (23-25, 28). NME was computed as \[ \Delta \text{Pdi}/\Delta \text{EMGdi} \].

Effort of the inspiratory muscles was quantified by calculating the esophageal pressure-time product (PTPes) and work of breathing (WOB) during inspiration. PTPes was calculated as the time integral of the difference between Pes and the recoil pressure of the chest wall, as described previously (29, 30). WOB was calculated...
as the integral of the product of $V_t$ and $P_{es}$ (i.e. the Campbell diagram) (31). $PT_P$ was partitioned in resistive, elastic and $P_{EEPi}$ components. A calculated theoretical value was used for the recoil pressure of the chest wall, as described previously (32). Effort of the expiratory muscles was quantified by calculating the gastric pressure-time product ($PT_{Pga}$) during expiration. $PT_{Pga}$ was calculated as the time integral of the rise in gastric pressure during expiration. Total $PT_P$ ($PT_{Ptot}$) for the respiratory muscles was calculated as $PT_{Pes} + PT_{Pga}$.

**Data analysis**

Data were analyzed on a breath-by-breath basis. For the period of mechanical ventilation, a 2-min period at the end was selected for analysis. The duration of the SBT differed per patient. Therefore, data were analyzed at seven points in time: the first and last minute and five periods of at least 1 min taken at equal time intervals in between.

Statistical analyses were performed using IBM SPSS Statistics version 22 (IBM Corp., Armonk, New York, USA). First, to analyze the effect of removing ventilator assist, a paired student t-test (PSV vs. first minute of the SBT) was performed for the different physiological variables per group (weaning failure and weaning success). Second, we used a linear mixed model design and applied restricted maximum likelihood estimation to analyze the effect of SBT duration (seven points from minute one to the last minute) and SBT group (weaning failure and weaning success). Linear mixed models are well suited for the analysis of repeated measures. In contrast to repeated measures two-way ANOVA it preserves more information contained in the data, they account for correlations between the repeated measurements within each subject and they can handle missing data (33). Full-factorial models were built with duration and group treated as fixed effects and subjects as a random effect. The model was run using the compound symmetry covariance type for the repeatedly measured outcomes. Bonferroni corrected pairwise comparisons based on the estimated marginal means were used as post-hoc tests. To analyze the effect of the SBT on blood gas values and hemodynamics a repeated measures two-way ANOVA was performed. For all tests, a two-tailed $p < 0.05$ was considered significant. Data are described as mean ± SEM, except as stated otherwise.
**Results**

Ten patients met the criteria for weaning failure, six patients failed the SBT after 30 ± 6.5 minutes (range 13 – 60 minutes) and four were reintubated within 48 hours after extubation. Eleven patients successfully completed the 60 minute SBT and remained extubated for at least 48 hours. Patient characteristics and ventilator settings at study inclusion are presented in Table 1. ΔEMGdi, CFdi and ΔPdi of one patient in the weaning success group were excluded from analysis due to dislocation of the catheter during SBT. From another patient in the failure group ΔEMGdi and CFdi were excluded from analysis due to electrode artefacts detected during offline signal analysis.

**Table 1.** Patient characteristics

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>Age (yr)</th>
<th>BMI (kg/m²)</th>
<th>Sex</th>
<th>Days MV</th>
<th>PSV level (cmH₂O)</th>
<th>NAVA level (cmH₂O/µV)</th>
<th>PEEP level (cmH₂O)</th>
<th>FiO₂</th>
<th>PaO₂/FiO₂ (mmHg)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Failure group</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1 Cardiac arrest</td>
<td>88</td>
<td>24</td>
<td>F</td>
<td>24</td>
<td>-</td>
<td>2.3</td>
<td>6</td>
<td>0.4</td>
<td>268</td>
</tr>
<tr>
<td>2 Pneumonia</td>
<td>64</td>
<td>22</td>
<td>F</td>
<td>49</td>
<td>10</td>
<td>-</td>
<td>6</td>
<td>0.3</td>
<td>328</td>
</tr>
<tr>
<td>3 Postoperative, CABG</td>
<td>72</td>
<td>27</td>
<td>F</td>
<td>83</td>
<td>10</td>
<td>-</td>
<td>8</td>
<td>0.4</td>
<td>194</td>
</tr>
<tr>
<td>4 Multitrauma</td>
<td>45</td>
<td>28</td>
<td>M</td>
<td>4</td>
<td>10</td>
<td>-</td>
<td>5</td>
<td>0.3</td>
<td>318</td>
</tr>
<tr>
<td>5 Exacerbation COPD</td>
<td>58</td>
<td>25</td>
<td>M</td>
<td>14</td>
<td>8</td>
<td>-</td>
<td>10</td>
<td>0.3</td>
<td>405</td>
</tr>
<tr>
<td>6 Postoperative, CABG</td>
<td>67</td>
<td>29</td>
<td>F</td>
<td>8</td>
<td>0</td>
<td>-</td>
<td>8</td>
<td>0.4</td>
<td>214</td>
</tr>
<tr>
<td>7 Postoperative, AAA</td>
<td>63</td>
<td>27</td>
<td>M</td>
<td>7</td>
<td>4</td>
<td>-</td>
<td>8</td>
<td>0.4</td>
<td>199</td>
</tr>
<tr>
<td>8 Postoperative, papillary muscle rupture</td>
<td>80</td>
<td>24</td>
<td>M</td>
<td>8</td>
<td>0</td>
<td>-</td>
<td>8</td>
<td>0.45</td>
<td>132</td>
</tr>
<tr>
<td>9 Guillain Barre</td>
<td>64</td>
<td>23</td>
<td>M</td>
<td>153</td>
<td>10</td>
<td>-</td>
<td>8</td>
<td>0.25</td>
<td>*</td>
</tr>
<tr>
<td>10 Cardiogenic shock</td>
<td>76</td>
<td>23</td>
<td>M</td>
<td>21</td>
<td>-</td>
<td>0.4</td>
<td>10</td>
<td>0.3</td>
<td>228</td>
</tr>
<tr>
<td><strong>Success group</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1 Tracheal stenosis (COPD)</td>
<td>69</td>
<td>25</td>
<td>M</td>
<td>7</td>
<td>14</td>
<td>-</td>
<td>3</td>
<td>0.45</td>
<td>190</td>
</tr>
<tr>
<td>2 Postoperative, aortoenteric fistula (COPD)</td>
<td>71</td>
<td>26</td>
<td>M</td>
<td>8</td>
<td>5</td>
<td>-</td>
<td>6</td>
<td>0.4</td>
<td>*</td>
</tr>
<tr>
<td>3 Sepsis</td>
<td>58</td>
<td>26</td>
<td>F</td>
<td>7</td>
<td>0</td>
<td>-</td>
<td>6</td>
<td>0.35</td>
<td>330</td>
</tr>
<tr>
<td>4 Postoperative, CABG</td>
<td>73</td>
<td>29</td>
<td>M</td>
<td>14</td>
<td>10</td>
<td>-</td>
<td>10</td>
<td>-</td>
<td>*</td>
</tr>
<tr>
<td>5 Cardiac arrest</td>
<td>76</td>
<td>29</td>
<td>F</td>
<td>17</td>
<td>6</td>
<td>-</td>
<td>6</td>
<td>0.45</td>
<td>248</td>
</tr>
<tr>
<td>6 Postoperative, AAA</td>
<td>82</td>
<td>28</td>
<td>M</td>
<td>5</td>
<td>9</td>
<td>-</td>
<td>5</td>
<td>0.45</td>
<td>258</td>
</tr>
<tr>
<td>7 Sepsis</td>
<td>73</td>
<td>29</td>
<td>M</td>
<td>24</td>
<td>8</td>
<td>-</td>
<td>6</td>
<td>0.25</td>
<td>306</td>
</tr>
<tr>
<td>8 Multitrauma</td>
<td>46</td>
<td>25</td>
<td>M</td>
<td>9</td>
<td>0</td>
<td>-</td>
<td>5</td>
<td>0.3</td>
<td>*</td>
</tr>
<tr>
<td>9 Sepsis</td>
<td>63</td>
<td>24</td>
<td>F</td>
<td>7</td>
<td>0</td>
<td>-</td>
<td>8</td>
<td>0.3</td>
<td>*</td>
</tr>
<tr>
<td>10 Pneumonia</td>
<td>60</td>
<td>21</td>
<td>M</td>
<td>12</td>
<td>6</td>
<td>-</td>
<td>6</td>
<td>0.35</td>
<td>291</td>
</tr>
<tr>
<td>11 Postoperative, CABG</td>
<td>78</td>
<td>30</td>
<td>M</td>
<td>3</td>
<td>4</td>
<td>-</td>
<td>10</td>
<td>0.4</td>
<td>358</td>
</tr>
</tbody>
</table>

* patient without arterial line. AAA, abdominal aortic aneurysm; CABG, coronary artery bypass graft; COPD, chronic obstructive pulmonary disease; F, female; M, male; MV, mechanical ventilation.
Ventilation, respiratory timing and mechanics

Variables of ventilation and respiratory time are listed in Table 2. Directly after the transition from PSV to spontaneous breathing there was a drop in $V_t$ in both groups, thereafter $V_t$ remained stable during the SBT. In both groups respiratory frequency and the ratio between respiratory frequency and $V_t$ (i.e. an index of rapid shallow breathing) increased after removal of ventilator assist. However, in the failure group there was a further increase in respiratory frequency and rapid shallow breathing during the SBT whereas this was not observed in the success group. There were no significant differences in $V_e$ over time and between groups.

Values of PEEPi, $C_{dyn \_L}$ and $R_{insp}$ are listed in Table 2. In the failure group PEEPi increased after removal of ventilator assist, but during the SBT there were no differences between both groups. Directly after the transition from PSV to spontaneous breathing $C_{dyn \_L}$ decreased in the failure group, whereas it remained stable in the success group. Removal of ventilator assist resulted in an increase in $R_{insp}$ in both groups. During the SBT no changes in or differences between groups for both $C_{dyn \_L}$ and $R_{insp}$ were found.

Respiratory muscle activity

Figure 1 shows the activity of the inspiratory muscle and expiratory muscles for both the weaning success and weaning failure group. Removal of ventilator assist caused an immediate increase in $\Delta P_{di}$, $\Delta P_{es}$ and $\Delta EMG_{di}$ in both groups, whereas $\Delta P_{ga \_exp}$ and NME of the diaphragm did not change in the first minute after the removal of inspiratory support.

During spontaneous breathing, no significant interactions were seen between SBT duration and group for $\Delta P_{di}$, $\Delta P_{es}$, $\Delta EMG_{di}$ and NME. However, due to a higher $\Delta EMG_{di}$, NME was lower in the weaning failure group compared to successfully extubated patients (Figure 1, panel D). In the failure group the activity of the expiratory muscles increased during spontaneous breathing, whereas it remained equal in the success group (Figure 1, panel E). The Gilbert index ($\Delta P_{ga} / \Delta P_{di}$) was higher in the success group compared to the failure group, indicating a greater contribution of the diaphragm to respiration.

$CF_{di}$, a measure of muscle fiber conduction velocity, decreased during the SBT in both the success group and failure group (significant main effect duration, $p = 0.002$) from $89 \pm 4$ Hz to $81 \pm 5$ Hz and from $93 \pm 2$ Hz to $88 \pm 2$ Hz, respectively. There was no significant interaction between the main effects of duration and group on $CF_{di}$ ($p = 0.874$).
Table 2. Variables of ventilation, respiratory timing and mechanics during PSV and the SBT for the failure group and success group

<table>
<thead>
<tr>
<th></th>
<th>PSV</th>
<th>SBT start</th>
<th>SBT end</th>
<th>p value main effect duration</th>
<th>p value main effect group</th>
<th>p value interaction duration * group</th>
</tr>
</thead>
<tbody>
<tr>
<td>VT (ml)</td>
<td>F 522 ± 49 *</td>
<td>367 ± 57</td>
<td>388 ± 65</td>
<td>0.866</td>
<td>0.715</td>
<td>0.424</td>
</tr>
<tr>
<td></td>
<td>S 466 ± 70 *</td>
<td>414 ± 62</td>
<td>413 ± 52</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Peak insp. flow (ml/s)</td>
<td>F 768 ± 72 *</td>
<td>619 ± 69</td>
<td>792 ± 106</td>
<td>&lt; 0.001</td>
<td>0.444</td>
<td>0.002</td>
</tr>
<tr>
<td></td>
<td>S 714 ± 74 *</td>
<td>637 ± 81</td>
<td>650 ± 78</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Freq. (breaths/min)</td>
<td>F 21.8 ± 2.8 *</td>
<td>26.2 ± 2.6</td>
<td>30.8 ± 2.6</td>
<td>0.007</td>
<td>0.772</td>
<td>0.012</td>
</tr>
<tr>
<td></td>
<td>S 24.9 ± 2.5 *</td>
<td>27.6 ± 2.8</td>
<td>28.0 ± 2.7</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>VTi (L/min)</td>
<td>F 10.9 ± 1.3</td>
<td>9.3 ± 1.5</td>
<td>11.5 ± 1.9</td>
<td>0.039</td>
<td>0.836</td>
<td>0.297</td>
</tr>
<tr>
<td></td>
<td>S 11.5 ± 1.5</td>
<td>11.1 ± 1.6</td>
<td>11.6 ± 1.4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ti (s)</td>
<td>F 0.99 ± 0.11</td>
<td>0.87 ± 0.08</td>
<td>0.70 ± 0.06</td>
<td>&lt; 0.001</td>
<td>0.101</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td></td>
<td>S 0.95 ± 0.07</td>
<td>0.96 ± 0.08</td>
<td>0.96 ± 0.10</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Te (s)</td>
<td>F 2.23 ± 0.36 *</td>
<td>1.78 ± 0.27</td>
<td>1.49 ± 0.18</td>
<td>0.122</td>
<td>0.679</td>
<td>0.099</td>
</tr>
<tr>
<td></td>
<td>S 1.76 ± 0.24 *</td>
<td>1.48 ± 0.19</td>
<td>1.47 ± 0.16</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ttot (s)</td>
<td>F 3.22 ± 0.45 *</td>
<td>2.65 ± 0.32</td>
<td>2.19 ± 0.23</td>
<td>0.024</td>
<td>0.814</td>
<td>0.020</td>
</tr>
<tr>
<td></td>
<td>S 2.71 ± 0.29 *</td>
<td>2.44 ± 0.25</td>
<td>2.43 ± 0.24</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ti/Ttot</td>
<td>F 0.32 ± 0.02</td>
<td>0.35 ± 0.02</td>
<td>0.33 ± 0.02</td>
<td>0.375</td>
<td>0.029</td>
<td>0.727</td>
</tr>
<tr>
<td></td>
<td>S 0.37 ± 0.02 *</td>
<td>0.41 ± 0.02</td>
<td>0.40 ± 0.02</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Freq./VTi (breaths/min/L)</td>
<td>F 46 ± 9 *</td>
<td>84 ± 13</td>
<td>101 ± 17</td>
<td>0.016</td>
<td>0.395</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>S 61 ± 9 *</td>
<td>78 ± 10</td>
<td>78 ± 10</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PEEPi (cmH2O)</td>
<td>F 2.3 ± 0.9 *</td>
<td>4.6 ± 1.2</td>
<td>5.0 ± 1.5</td>
<td>0.654</td>
<td>0.250</td>
<td>0.802</td>
</tr>
<tr>
<td></td>
<td>S 2.0 ± 0.5</td>
<td>2.8 ± 0.8</td>
<td>3.4 ± 0.9</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CdynL (ml/cmH2O)</td>
<td>F 48.9 ± 8.0 *</td>
<td>33.6 ± 5.3</td>
<td>31.7 ± 7.6</td>
<td>0.508</td>
<td>0.664</td>
<td>0.691</td>
</tr>
<tr>
<td></td>
<td>S 41.8 ± 4.9</td>
<td>40.8 ± 9.0</td>
<td>35.9 ± 7.4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rinsp (cmH2O/L/s)</td>
<td>F 8.2 ± 2.5 *</td>
<td>15.3 ± 3.0</td>
<td>16.0 ± 2.7</td>
<td>0.926</td>
<td>0.909</td>
<td>0.545</td>
</tr>
<tr>
<td></td>
<td>S 7.3 ± 1.1 *</td>
<td>14.4 ± 2.5</td>
<td>15.9 ± 3.3</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Note that only data at start and end of the SBT is shown and not the time intervals in between. * p < 0.05 PSV vs. SBT start (student t-test). p values in table reflect results during the SBT (linear mixed model). CdynL, dynamic lung compliance; PEEPi, positive end-expiratory pressure; PSV, pressure support ventilation; F, weaning failure group; S, weaning success group; Rinsp, inspiratory resistance; SBT, spontaneous breathing trial; Te, expiratory time; Ti, inspiratory time; Ttot; total respiratory cycle time; VTi, minute ventilation; VT, tidal volume.
Figure 1. ΔPdi, ΔPes, ΔEMGdi, NME, ΔPga_{exp} and the Gilbert index during PSV and the SBT for the weaning success and weaning failure group. After removal of ventilator assist ΔPdi, ΔPes and ΔEMGdi increased in both groups (* p < 0.05 PSV vs. start SBT). During the SBT there was a significant main effect of duration (p = 0.047) on ΔPes (panel B), a significant main effect of duration (p = 0.039) and group (p = 0.047) on ΔEMGdi (panel C), a significant main effect of group (p = 0.033) on NME (panel D), a significant main effect of duration (p = 0.013) on ΔPga_{exp} (panel E) and a significant main effect of group (p = 0.020) on the Gilbert index (panel F). * p < 0.05 Bonferroni corrected pairwise post-hoc comparisons. EMGdi, diaphragm electromyography; NME, neuromechanical efficiency; Pdi, transdiaphragmatic pressure; Pes, esophageal pressure; Pga, gastric pressure; ΔPga_{exp}, expiratory rise in gastric pressure; PSV, pressure support ventilation; SBT, spontaneous breathing trial.
Respiratory muscle effort

Figure 2 (panel A) shows the inspiratory muscle effort. PTPes was equal during PSV for both groups. There was initial and parallel significant increase in PTPes in both groups after removal of ventilator assist. During the SBT there were no effects of duration or group on PTPes. In addition, there were no differences between the PEEPi, elastic and resistive components of PTPes.

Figure 2 (panel B) shows the expiratory muscle effort. The transition from PSV to spontaneous breathing had no effect on PTPga. However, during the SBT there was an significant interaction between duration of the SBT and group, indicating that the increase in PTPga in the failure group was significantly higher than in the success group. Figure 3 shows an representative example of increasing expiratory rise in Pga during the SBT. Moreover, as can be seen in Figure 4, in the failure group the contribution of the expiratory muscles to total respiratory muscle effort increased from $15 \pm 4\%$ to $26 \pm 4\%$ during spontaneous breathing, whereas there were no changes in the success group.
Inspiratory WOB during PSV was 9.8 ± 2.7 J/min in the success group and 8.8 ± 2.4 J/min in the failure group, which increased to 14.4 ± 3.3 J/min (p = 0.003) and 15.7 ± 3.9 J/min (p < 0.001), respectively, after removal of ventilator assist. Although WOB at the end of the SBT was 17.1 ± 3.8 J/min for the success group and 23.7 ± 9.5 J/min for the failure group, these differences did not reach statistical significance.

Arterial blood gas measurements
In Table 3, the blood gas values are presented. There were significant main effects of time point and group on PaCO₂, such that PaCO₂ increased over time during the SBT and was higher in the failure group. However, the interaction between the effects of time point and group was not significant, indicating that the increase in PaCO₂ was not different between both groups.

Table 3. Blood gas values and hemodynamic variables before and after the SBT for the failure group and the success group.

<table>
<thead>
<tr>
<th></th>
<th>PSV</th>
<th>SBT end</th>
<th>p value main effect time point</th>
<th>p value main effect group</th>
<th>p value interaction time point * group</th>
</tr>
</thead>
<tbody>
<tr>
<td>PaO₂ (mmHg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>95 ± 9</td>
<td>103 ± 16</td>
<td>0.735</td>
<td>0.930</td>
<td>0.532</td>
</tr>
<tr>
<td>S</td>
<td>102 ± 7</td>
<td>99 ± 9</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PaCO₂ (mmHg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>46 ± 3</td>
<td>50 ± 4</td>
<td>0.021</td>
<td>0.046</td>
<td>0.187</td>
</tr>
<tr>
<td>S</td>
<td>38 ± 1</td>
<td>40 ± 2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pH</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>7.42 ± 0.02</td>
<td>7.40 ± 0.02</td>
<td>0.013</td>
<td>0.228</td>
<td>0.471</td>
</tr>
<tr>
<td>S</td>
<td>7.45 ± 0.01</td>
<td>7.44 ± 0.01</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MAP (mmHg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>87 ± 7</td>
<td>89 ± 8</td>
<td>0.903</td>
<td>0.579</td>
<td>0.281</td>
</tr>
<tr>
<td>S</td>
<td>96 ± 10</td>
<td>94 ± 9</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heart rate</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>84 ± 6</td>
<td>87 ± 8</td>
<td>0.228</td>
<td>0.571</td>
<td>0.505</td>
</tr>
<tr>
<td>S</td>
<td>81 ± 4</td>
<td>82 ± 3</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

MAP, mean arterial pressure; PSV, pressure support ventilation; F, weaning failure; S, weaning success; SBT, spontaneous breathing trial.

Discussion
The present study is the first that provides an in depth analysis of the recruitment and effort of respiratory muscles during inspiration and expiration in a heterogeneous group of weaning patients. We found that after removal of ventilator assist, patients
Inspiratory and expiratory muscle effort during weaning

Figure 3. Individual example of increasing expiratory rise in Pga during the SBT. Note that the increase in gastric pressure during expiration caused by the expiratory muscle exceeds the increase in gastric pressure during inspiration caused by the diaphragm. PSV, pressure support ventilation; Pes, esophageal pressure; Pga, gastric pressure; SBT, spontaneous breathing trial.

Figure 4. The contribution of the expiratory muscles to total respiratory muscle effort during PSV and the SBT for the weaning success and weaning failure group. During the SBT there was a significant main effect of duration (p = 0.011) and group (p = 0.038) and a significant interaction between the effects of duration and group (p = 0.028) on PTPga/PTPtot. * p < 0.05 Bonferroni corrected pairwise post-hoc comparison. PTPga, gastric pressure-time product; PTPtot, total pressure-time product; SBT, spontaneous breathing trial.
who fail a trial of spontaneous breathing progressively developed an increase in expiratory muscle effort up to 26% of total respiratory muscle effort at the end of the trial, while expiratory muscle effort in the success group remained stable. Furthermore, the failure group has a lower neuro-mechanical efficiency of the diaphragm on PSV and during SBT compared to successful weaning patients.

**Expiratory muscle effort**

Respiratory muscle effort during the breathing cycle can be quantified using the pressure-time product, i.e. the time integral of the difference between esophageal pressure and the recoil pressure of the chest wall (29, 30). In experimental studies, changes in PTPes have been shown to reflect a comparable change in diaphragmatic energy expenditure (34, 35). After removal of ventilator assist we found an increase in PTPes, but no differences were found between the success group and failure group. However, under conditions of increased loading and/or impaired diaphragm function, expiratory muscles may be recruited during expiration (18). Indeed, in the present study expiratory muscle activity, as indicated by an expiratory rise in Pga, increased in the failure group during the SBT. This is consistent with previous findings in patients with COPD (19, 20). Expiratory muscle recruitment will increase the energy expenditure of respiration. Notably, in healthy subjects, it was demonstrated that expiratory pressure generation is a significant contributor to the perception of dyspnoea during exercise (36). Therefore, the expiratory component should be taken into account when calculating total respiratory muscle effort. In the present study, we introduce the expiratory pressure-time product, i.e. the time integral of the rise in gastric pressure during expiration and calculated the contribution of the expiratory muscles to total respiratory muscle effort. Remarkably, in the weaning failure group, the contribution of the expiratory muscles to total respiratory muscle effort increased up to 26%, whereas it remained stable in successfully extubated patients.

It has been reasoned that the goal of expiratory muscle recruitment is to assist the inspiratory muscles by decreasing end-expiratory lung volume (18, 37). However, PEEPi (corrected for the expiratory rise in Pga) in the current study, a surrogate of end-expiratory lung volume, was not lower in the failure group compared to the success group. On the contrary, PEEPi tended to be higher in the failure group. In addition, there were no improvements in NME of the diaphragm during the SBT in the failure group that would support a beneficial effect of expiratory muscle recruitment on inspiratory muscle function. In COPD patients it has also been demonstrated that phasic contraction of the expiratory muscles does not increase NME of the diaphragm (37). Our findings raise the possibility that expiratory muscle recruitment might not support the inspiratory muscles, but instead contributed to weaning.
Inspiratory and expiratory muscle effort during weaning failure. However, one could also argue that NME and PEEPi would have been worse at the end of the SBT in the failure group if expiratory muscles would not have recruited. Whether increased expiratory muscle effort contributes to weaning failure or prevents higher inspiratory muscle effort should be investigated in subsequent studies.

**Neuromechanical efficiency of the diaphragm**

As expected, after removal of ventilator assist both ΔPdi and ΔEMGdi increased in the success group and the failure group. There was no difference between groups for the increase in ΔPdi and ΔEMGdi, but there was a clear baseline difference in ΔEMGdi before and during the SBT. Consequently, NME of the diaphragm was constantly lower in patients who failed a trial of spontaneous breathing (± 0.4 cmH₂O/µV) compared to the success group (± 0.7 cmH₂O/µV). In healthy subjects NME is approximately 1.4 cmH₂O/µV (15), thus NME is approximately 30% and 50% of normal in patients who fail a SBT and successfully extubated patients, respectively. To our knowledge, there are no other studies that continuously measured NME during a T-tube trial using Pdi and EMGdi. Previously, NME has been calculated intermittently during a 30-minute weaning trial as the decrease in airway pressure divided by ΔEMGdi during an inspiratory occlusion (38). In the latter study, the weaning trial was performed with 5 cm H₂O of continuous positive airway pressure (CPAP). The addition of 5 cm H₂O of PEEP can decrease work of breathing by as much as 40% in ventilated patients (30). Despite different methods of weaning trials and calculation of NME, the finding of reduced ability of the diaphragm to convert neural respiratory drive into inspiratory pressure is consistent in patients who fail a weaning trial.

Undoubtedly the reduced NME we found in both groups is partly due to weakness of the diaphragm. Over the last years evidence of diaphragm weakness in critically ill patients has accumulated (7, 9-13). However, the pressure-generating capacity of the diaphragm also depends on muscle fatigue, and the force-velocity and force-length relations. These factors may play an important role during a spontaneous breathing trial. We did not find any evidence for short-lasting fatigue of the diaphragm, as expressed by center frequency of the diaphragm EMG. In addition, previously it was demonstrated that a one hour SBT did not cause long-lasting fatigue of the diaphragm (39). Increased end-expiratory lung volume may have placed the diaphragm in a less favourable length-tension curve. An increase in lung volume from functional residual capacity to total lung capacity reduces Pdi by 60% for a given ΔEMGdi (40). Although we did not measure end-expiratory lung volume, corrected PEEPi was not significantly higher in the failure group, suggesting no major differences in lung volume. Overall, the reduced neuromechanically efficiency in the failure group may
largely be explained by diaphragmatic weakness, but we cannot completely rule out impaired respiratory mechanics.

In the present study, increased respiratory muscle effort (including expiratory muscle effort) and reduced neuromechanical efficiency of the diaphragm appear to play a role in failure to wean from mechanical ventilation. However, it is important to note that reduced respiratory muscle capacity and increased respiratory load are not the only causes of failure to wean from mechanical ventilation. Cardiac dysfunction, cognitive dysfunction and metabolic disorders are recognized causes for weaning failure as well (5, 6) and may have contributed to weaning failure in the current study.

**Limitations**

In the present study, raw EMGdi signals were recorded from multiple esophageal electrodes and processed according to the method of Sinderby and colleagues using an in-house developed algorithm (21, 23-25). Today many studies record electrical activity of the diaphragm not in its raw format (EMGdi), but as a processed signal via the neurally adjusted ventilatory assist module of the Servo-i ventilator. Differences between algorithms to process EMGdi can lead to differences in absolute ΔEMGdi. Therefore, values of ΔEMGdi (and NME) obtained in our study cannot be directly compared to values obtained with recordings of electrical activity of the diaphragm from the Servo-I ventilator.

We did not record EMG of the abdominal muscles in the current study to detect expiratory muscle recruitment. However a high correlation has been demonstrated between the increase in gastric pressure during expiration and electrical activity of the abdominal wall muscles (41).

In conclusion, the expiratory muscles significantly contribute to respiratory muscle effort in a mixed group of critically ill patients who fail a trial of spontaneous breathing. Therefore, the expiratory pressure-time product should be measured when estimating energy expenditure of the respiratory muscles during weaning from mechanical ventilation. In addition, our findings confirm that impaired pressure-generating capacity of the diaphragm, regardless of its origin, plays a role in failure to wean from mechanical ventilation.
Inspiratory and expiratory muscle effort during weaning

References


Chapter 5

Functional assessment of the diaphragm by ultrasonic deformation imaging during inspiratory loading

Eline Oppersma, Nima Hatam, Jonne Doorduin, Johannes G. van der Hoeven, Gernot Marx, Andreas Goetzenich, Sebastian Fritsch, Leo M.A. Heunks, Christian B. Bruells

Accepted for publication in J Appl Physiol
Abstract

Background: Assessment of diaphragmatic effort is challenging especially in critically ill patients in the phase of weaning. Fractional thickening during inspiration assessed by ultrasound has been used to estimate diaphragm effort. It is unknown whether more sophisticated ultrasound techniques such as speckle tracking are superior in the quantification of inspiratory effort. This study evaluates the validity of speckle tracking ultrasound to quantify diaphragm contractility.

Methods: Thirteen healthy volunteers underwent a randomized stepwise threshold loading protocol of 0 to 50% of the maximal inspiratory pressure. Electric activity of the diaphragm and transdiaphragmatic pressures were recorded. Speckle tracking ultrasound was used to assess strain and strain rate as measures of diaphragm tissue deformation and deformation velocity, respectively. Fractional thickening was assessed by measurement of diaphragm thickness at end-inspiration and end-expiration.

Results: Strain and strain rate increased with progressive loading of the diaphragm. Both strain and strain rate were highly correlated to transdiaphragmatic pressure (strain $R^2=0.72$; strain rate $R^2=0.80$) and diaphragm electric activity (strain $R^2=0.60$; strain rate $R^2=0.66$).

Conclusion: Speckle tracking ultrasound is superior to conventional ultrasound techniques to estimate diaphragm contractility under inspiratory threshold loading.
Introduction

Under physiological conditions, the pressure developed by the inspiratory muscles is only ±5% of maximum inspiratory pressure (1). However, under pathological conditions, such as an acute exacerbation of chronic obstructive pulmonary disease or a failed trial of weaning from mechanical ventilation, the load imposed on the respiratory muscles increases considerably (2, 3). Excessive inspiratory muscle loading may result in fatigue or injury of the diaphragm (4-6). Accordingly, in selected patients evaluating respiratory muscle effort may be of clinical relevance (7, 8). Today, measurement of transdiaphragmatic pressure (Pdi) using esophageal and gastric balloons is the gold standard to assess effort of the diaphragm. However, this technique is invasive, requires expertise and interpretation may be complex (7, 9).

Diaphragmatic function has been studied by B-mode and M-mode ultrasound (10-12). Fractional thickening (FT) of the diaphragm has been used in previous studies to quantify effort of the diaphragm (11, 13, 14). However, Goligher et al. reported low correlations ($R^2=0.28$) in healthy subjects between Pdi versus diaphragmatic thickening fraction (15), indicating limited validity of FT to quantify diaphragm effort.

Two-dimensional deformation ultrasound or speckle tracking (ST) ultrasound is an innovative ultrasound technique enabling distinct assessment of muscle function (16). The grey value pattern in ultrasound images remains relatively constant for any small region in muscle tissue, this is called a speckle. In the speckle tracking technique, a defined cluster of speckles is tracked from one frame to another during a contractile cycle. This enables the angle-independent, two-dimensional quantification of the percentage of deformation (strain, %) and deformation velocity (strain rate, sec$^{-1}$). For readers with additional interest in basic deformation imaging methodology we may humbly refer to the references of Collier et al and Smiseth et al (17, 18).

Speckle tracking echocardiography has become a popular tool for both research and clinical purposes (16, 19-21). Previously, we have demonstrated the feasibility of ST of the diaphragm during respiratory muscle unloading with non-invasive ventilation (22). ST has not been validated as a measure of diaphragm contractility using Pdi as a gold standard. We hypothesize that strain and strain rate, obtained by ST, can be used to quantify diaphragm contractility during inspiratory loading and this study aims to investigate the validity of these parameters. Part of this work has previously been presented at the international conference of the European Respiratory Society (23).
Materials and methods

Subjects
We enrolled 15 healthy volunteers with a body mass index less than 25 kg/m². The study was conducted at the Radboud university medical center and the protocol was approved by the local ethics review committee and conducted in accordance with the Declaration of Helsinki and its later amendments. All subjects gave their written informed consent. Pre-existent neuromuscular disorders or lung diseases were defined as exclusion criteria.

Measurements
A multi-electrode esophageal catheter with two balloons (NeuroVent Research Inc, Toronto, Canada) was inserted and the balloons were inflated with air as described previously (24, 25). The flow, electric activity of the diaphragm (EAdi), esophageal pressure (Pes), and gastric pressure (Pga) were recorded continuously (25). EAdi signals were amplified and digitized (Porti 16, 22 bits, 71.5 nV/least significant bit, TMSi; The Netherlands) at a sampling frequency of 2 kHz. Pressure signals and flow were digitized (Porti 16, 22 bits, 1.4 µV/least significant bit, TMSi; The Netherlands) at a sampling frequency of 2 kHz. Data were stored and buffered on an external drive for offline analysis. Transdiaphragmatic pressure (Pdi) was calculated as Pes subtracted from Pga. Tidal volume (Vₜ) was obtained by digital integration of the flow signal. Diaphragm ultrasound was performed using a 9 MHz linear transducer with a Vivid E 9TM ultrasound machine (General Electric Healthcare, Horton, Norway).

Study protocol
The protocol starts with the measurement of maximum inspiratory pressure (MIP). The mean mouth pressure (Pmo) during sustained maximum inspiration for 1 second as recommended by the ATS/ERS statement on respiratory muscle testing (9) against a closed valve at functional residual capacity is defined as the MIP. The maneuver is repeated at least five times, until three reproducible efforts, with less than 10% variance, are obtained. Subjects were seated in upright position.

Inspiratory loading
An in-house developed inspiratory threshold apparatus, modified from Chen et al. (26) was used to perform negative pressure threshold loading. In short, the device consisted of a cylindrical adjustable pressure chamber, which was connected to a non-rebreathing valve. The negative pressure was generated by a powerful commercially available vacuum cleaner. Pressure in the chamber was measured continuously using
a differential pressure transducer (range ± 375 mmHg; Freescale, USA). The dead space of the device can be estimated around 600 ml. Subjects were seated in upright position with uncast abdomen, breathing through a mouthpiece while wearing a nose clip. Inspiratory loading of 0, 10, 20, 30, 40 and 50% of MIP was applied in random order. Every loading task was applied for 3 minutes, alternated with 5 minutes of unloaded breathing. During the loading tasks, EAdi, Pes, Pga, Pmo and flow were recorded continuously (Figure 1).

Ultrasound recording
The ultrasound transducer was positioned in the right anterior axillary line longitudinal to the body axis (between the 9th-11th intercostal space) (Figure 2 upper panel). We chose the strongly longitudinal approach versus the individual intercostal space to reduce angle-dependence of the measurements. The hemi-diaphragm is thereby displayed above the liver as a central less-echogenic layer between the peritoneal and pleural echogenic layer (Figure 2 upper panel). The region of interest (ROI) was positioned as described below during the offline data analysis (Figure 2 lower panel). This probe position was marked on the skin for the purpose of standardization (14). Ultrasound recordings of the diaphragm were made at the final minute of every inspiratory loading task. A 10 second recording with the highest possible frame rate was used for offline analysis. Strain describes the relative change in length between an initial reference state (L₀) and compressed/shortened state (L). The conventional strain is defined as: \( \varepsilon = \frac{L - L_0}{L_0} \). Positive strain means stretching, whereas negative strain means shortening. An increase of strain, as described in the following data presentation, refers to a more negative value of strain, thus an increase of shortening; e.g. -10 to -15 % corresponds to an increase of strain (or shortening).

Strain rate indicates the rate of deformation as follows: \( \varepsilon' = \frac{d\varepsilon}{dt} \). Strain rate is an instantaneous measurement not requiring a relation to a reference state.

Data analysis
EAdi, Pes, Pga, Pdi, Pmo and flow were analyzed offline using algorithms developed in Matlab R2013a (The Mathworks, Natick, MA). EAdi refers to a method using a standard electrode, acquisition, and analysis system to overcome signal filtering and processing effects when quantifying the diaphragm, as described previously (25). Pes at end-expiration was calculated as a measure of lung volume, and corrected for active expiration (27). The recorded ultrasound data were analyzed offline with the speckle tracking technique using the 2D – Strain modality of EchoPac®’s Q-analysis tool (software version BT 12, General Electric Healthcare, Horton, Norway).
Figure 1. The figure shows a representative tracing of EAdi, flow, Pdi, Pes, Pga and Pmo during inspiratory threshold loading (20% of maximal inspiratory pressure, random subject). Shaded areas are inspiration. EAdi, electrical activity of the diaphragm; Pdi, transdiaphragmatic pressure; Pes, esophageal pressure; Pga, gastric pressure; Pmo, mouth pressure.
Figure 2. Upper panel (A): B-mode picture of a scanned diaphragm area between chest wall and liver. The arrows indicate the diaphragm between hyperechoic lines, which are the equivalent to the border between diaphragm/pleura and peritoneum. Note the grey “dots” inside the diaphragm (“speckles”). Lower panel (B): region of interest tracked by the software.
Although the GE software is designed to trigger by the ECG signal by default, the cine-loops were adjusted to one entire breathing cycle based upon simultaneous recorded respiratory curves. The software allows for manually moving the loop to the desired cycle resulting in an adequate measurement, independent of the cardiac cycle. The ROI was placed at the lower echogenic line (peritoneal line) to the upper echogenic line (pleural line) starting at the right side of the sector (cranial) and ending at the left sector side (caudal) with approximately 5 to 7 points (Figure 2 lower panel). In some loops, especially in the higher loading steps (40% and 50% MIP), several attempts of ROI placement were necessary to achieve proper tracking. Here, fewer points with a narrower ROI had to be selected to allow adequate tracking. In general, these offline measurements can be finished in several minutes.

FT was also calculated from the ultrasound images. In the 2D image, diaphragmatic thickness at end-expiration and end-inspiration was measured at the same point following the longitudinal downward motion of the respective location. FT was measured at minimum in three different breaths and calculated by the following equation: (thickness at inspiration - thickness at expiration)/thickness at expiration. The quality of the recorded loops varied in some volunteers depending on the load and general movement of the thorax especially in high loading steps. Loops were discarded from the analysis if no sufficient border tracking could be developed.

**Statistical analysis**

Values are presented as mean ± SD, and p<0.05 was considered significant. Statistical analyses were performed with SPSS 21.0 (SPSS, Chicago, IL). Repeated measures one-way ANOVA was used to test the effect of inspiratory loading on EAdi, Pes at end-expiration, Pdi, strain, strain rate and FT. Correlations between EAdi, Pdi, FT and strain and strain rate were assessed using repeated observations correlation (28, 29). This method accounts for multiple measurements within subjects, by removing the differences between subjects and looking only at changes within subjects.

**Results**

**Subjects**

All included subjects (baseline characteristics: male/female, 7/8; age, 21.3 ±2.3 yr; 21.6 ±1.7 kg/m²) completed the protocol without adverse effects. Two data sets were lost from analysis due to technical issues (file damage). Mean MIP value for the group was 100 ± 32 cmH₂O.
Table 1. Response in tidal volume and respiratory frequency to the different loading steps. Data are shown as means and standard deviation for all subjects. P-value is given of repeated measures one-way ANOVA.

<table>
<thead>
<tr>
<th>Inspiratory loading (% MIP)</th>
<th>0</th>
<th>10</th>
<th>20</th>
<th>30</th>
<th>40</th>
<th>50</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>VT (ml)</td>
<td>1076 ± 350</td>
<td>1183 ± 427</td>
<td>1147 ± 425</td>
<td>1053 ± 346</td>
<td>1049 ± 418</td>
<td>936 ± 329</td>
<td>0.161</td>
</tr>
<tr>
<td>Resp. frequency (breaths/min)</td>
<td>13.5 ± 4.2</td>
<td>15.3 ± 4.4</td>
<td>15.6 ± 5.4</td>
<td>16.7 ± 6.1</td>
<td>16.1 ± 5.7</td>
<td>17.3 ± 5.7</td>
<td>0.133</td>
</tr>
</tbody>
</table>

MIP, maximal inspiratory pressure; VT, tidal volume

Table 2. Thickness of the diaphragm at end-expiration, end-inspiration and thickening fraction. Data are shown as means and standard deviation for all subjects. The bottom line shows the values for the repeatability coefficient of the measurements.

<table>
<thead>
<tr>
<th>Inspiratory loading (% MIP)</th>
<th>End-expiration thickness (mm)</th>
<th>End-inspiration thickness (mm)</th>
<th>Thickening fraction (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>2.4 ± 0.7</td>
<td>3.8 ± 1.2</td>
<td>59.9 ± 32.1</td>
</tr>
<tr>
<td>10</td>
<td>2.4 ± 0.6</td>
<td>4.0 ± 1.5</td>
<td>67.9 ± 35.0</td>
</tr>
<tr>
<td>20</td>
<td>2.6 ± 0.5</td>
<td>4.1 ± 1.7</td>
<td>55.0 ± 39.2</td>
</tr>
<tr>
<td>30</td>
<td>2.6 ± 0.7</td>
<td>3.9 ± 1.8</td>
<td>48.9 ± 34.7</td>
</tr>
<tr>
<td>40</td>
<td>2.5 ± 0.6</td>
<td>4.0 ± 1.4</td>
<td>61.5 ± 48.1</td>
</tr>
<tr>
<td>50</td>
<td>2.4 ± 0.7</td>
<td>3.6 ± 1.4</td>
<td>50.6 ± 46.7</td>
</tr>
</tbody>
</table>

Repeatability coefficient 1.07 mm 2.54 mm 85.2%

MIP, maximal inspiratory pressure

Physiological measurements
Mean mouth pressure for all subjects was -1.2 ±0.4, -13.7 ±4.6, -26.3 ±7.6, -37.9 ±11.7, -49.9 ±14.3 and -59.7 ±16.9 cmH₂O for 0 to 50% inspiratory loading respectively. The stepwise increase of inspiratory loading resulted in an increase in both Pdi and EAdi (Figure 3). Pdi increased from 14.3 ±5.9 cmH₂O at unloaded breathing via the mouthpiece to 60.8 ±24.4 cmH₂O at 50% loading. Likewise, EAdi increased from 20.3 ±11.3 µV at zero loading to 66 ±23.2 µV at 50% loading. Pes at end-expiration decreased from -3.7 ± 3.7 cmH₂O at zero loading to -6.0 ± 5.3 cmH₂O at 50% loading (p=0.03). There were no changes in VT and respiratory frequency during the different inspiratory loading steps (Table 1).
Figure 3. Upper 4 panels: mean with SD of EAdi, Pdi, strain and strain rate as function of inspiratory loading (%MIP) for all subjects. Repeated measures one-way ANOVA showed a significant effect of increased loading on all variables (p<0.001). Lower 2 panels: individual data of all subjects, strain and strain rate as function of inspiratory loading (%MIP). EAdi, electrical activity of the diaphragm; MIP, maximal inspiratory pressure; Pdi, transdiaphragmatic pressure.
Assessment of diaphragm function by ultrasonic deformation imaging

Ultrasound assessment
In all assessable datasets, a ROI could be tracked in the offline ultrasound analysis during diaphragm contraction and relaxation. Table 2 shows the thickness of the diaphragm at end-expiration, end-inspiration and thickening fraction, as well as the repeatability coefficients. Repeated measures one-way ANOVA showed that with increasing load both strain and strain rate increased (p<0.001). Strain increased from -22 ±7.6% at zero loading to -41.5 ±10.1 at 50% loading. Consistent with strain, strain rate increased from -0.48 ±0.2 s\(^{-1}\) at zero loading to -1.5 ±0.7 s\(^{-1}\) at 50% loading (Figure 3).

Strain and strain rate were both significantly correlated with EAdi and Pdi. Strain versus EAdi showed a correlation of \(R^2=0.60\) (p<0.0001), while strain versus Pdi data showed a correlation of \(R^2=0.72\) (p<0.0001). Correlations between strain rate and Pdi (\(R^2=0.80\), p<0.0001) and between strain rate and EAdi (\(R^2=0.66\), p<0.0001) were even higher than reported for strain (Figure 4).

Figure 4. Correlation of Pdi and EAdi vs. strain and strain rate for all subjects during inspiratory loading from 0-50%. EAdi, electrical activity of the diaphragm; Pdi, transdiaphragmatic pressure.
Diaphragm thickness during zero loading was 2.4 ±0.7 mm and 3.8 ±1.2 mm during end-expiration and end-inspiration, respectively, and did not change during inspiratory loading (Table 2). Consequently, FT was not affected by incremental inspiratory loading (p=0.70). Also, no significant correlations between EAdi and FT (p=0.790) and between Pdi and FT (p=0.495) as well as between FT and strain (p=0.654) and strain rate (p=0.364) were found.

Discussion

The present study is the first to evaluate speckle tracking ultrasound of the diaphragm during inspiratory muscle loading. We found that the ST parameters strain and strain-rate are highly correlated with Pdi, the gold standard for diaphragmatic contractility, and also with EAdi. Strain rate had the highest correlation with Pdi and EAdi. Furthermore, ST proved to be superior to diaphragm fractional thickening as assessed by conventional ultrasound to quantify diaphragmatic effort.

Validation of the physiological model

Two methods for loading of the diaphragm have been described in the literature: inspiratory threshold loading and inspiratory resistive loading. The load imposed by the latter method is highly dependent on the inspiratory flow generated by the subject; low flow will result in minimal loading. We used a threshold load, as this may best reflect loading in intensive care patients where elastic properties of the respiratory system are increased due to pulmonary edema, chest-wall edema and pleural fluid. Our set up for inspiratory threshold loading was modified from Chen et al (26). Figure 1 shows a representative example of one subject at loading of 10% of MIP. It should be noted that at 0% inspiratory loading Pdi, EAdi, V_t and TF were higher than expected for a healthy subject (25). This is most likely the result of the additional resistance and instrumental dead space imposed by the experimental device. As expected, increasing the inspiratory load as a percentage of MIP, resulted in an increase in Pdi (Figure 3).

Speckle tracking ultrasound of the diaphragm

Tracking of unique greyscale scatter patterns, ‘speckles’, is one of the most useful tools in cardiac imaging, to assess cardiac function and is closely correlated to contractile myocardial function and outcome (16, 21, 30, 31). These speckle patterns are the ultrasonic correlate of interferences of ultrasound inside the tissue, i.e. different tissue patterns (muscle fibers, epimysium, etc.) and their movement towards each other.
Movement of speckle patterns reflects myofibrils/muscle tissue during its contraction, although speckles do not represent specific single myofibrils. It is important to mention that strain and strain rate approximate contractile function, but are not equal to contraction (32, 33).

Software tools to derive deformation data out of 2- or 3-dimensional ultrasonic cine loops are usually designed to track speckles in between endomyocard and epimyocard as hyperechoic leading structures. The parameters strain and strain rate define different, but load dependent variables: muscle deformation (strain) and deformation velocity (strain rate) can be inferred. Importantly, in contrast to conventional ultrasound techniques this measure is probe angle independent, which is of fundamental importance if the diaphragm is investigated. An important advantage of ST is, compared to conventional ultrasound, that the ST software recognizes the same region of the diaphragm. A limitation is that defining ROIs and the software calculations are based on an algorithm patented by GE, which is not open to the public.

Within the current study, we did not assess intra- and interobserver variability. This has been extensively evaluated by Orde et al (34). That study demonstrated that ST of the right diaphragm is feasible and reproducible (34). Diaphragm images were recorded from the end of expiration through the end of inspiration at 60% maximal inspiratory capacity. The current study is the first to demonstrate that diaphragm strain and strain rate are highly correlated with Pdi, the gold standard for diaphragm effort. In addition, a high correlation was found between these two ST measures and EAdi.

**Fractional thickening during inspiratory loading**

Diaphragm FT during inspiration as a measure for loading has been studied previously (15, 35-37). Umbrello et al. found that diaphragm thickening is a reliable indicator of respiratory effort (35) and Vivier et al. found as well a parallel decrease in TF and the diaphragmatic pressure-time product per breath during noninvasive ventilation with increasing levels of support (37). Goligher et al. (15) reported a rather low but significant correlation (R² 0.3; P<0.01) for FT versus both Pdi and EAdi in healthy subjects (N=5) (15). In the current study no significant correlation between FT and Pdi or EAdi was found. To evaluate if insufficient ultrasound training in our investigators contributed to this discrepancy, the repeatability coefficient of thickness at end-expiration was calculated, as this measure is unaffected by our loading protocol (Table 2). This demonstrates that the relatively high variation in FT in the present study is mainly due to the variation in thickness at end-inspiration, but also the repeatability at end-expiration in our study was relatively high compared to the study by Goligher et al. (15) (1.07 mm vs. 0.2 mm in the current study and the Goligher study respectively).
Another possible explanation for the apparent discrepancy with Golighers study is the difference in inspiratory loading protocol used. In their study, thickness was measured at different lung volumes, whereas in our study, inspiratory threshold loading was imposed, while volume was kept more or less constant (Table 1) which may have important implications. Lastly, the current study is a single point study and not a follow-up study where patients are ventilated; our population consists of healthy, not ventilated subjects, which also may account for the discrepancy with Golighers study. In a study by Cohn (38) healthy subjects were instructed to target specific lung volumes up to total lung capacity. A non-linear relationship was found for FT versus lung volume (polynomial equation was calculated with a R2 of 0.99). As subjects were instructed to keep the glottis open, the diaphragm was active at the targeted lung volumes, thus changes in diaphragm thickness resulted from changes in volume and pressure. When thickness is measured at different lung volumes with closed glottis (diaphragm relaxed), volume affects diaphragm thickness only at lung volume >50% of vital capacity (15). Finally, Ueki (39) measured diaphragm during maximal inspiratory effort against a closed valve (isovolumetric). They reported a strong correlation between maximum inspiratory pressure and diaphragm FT (R² = 0.67). The relationship between isometric inspiratory pressure and FT was not systematically studied.

Apparently, the relationship between diaphragm thickness and effort (Pdi, EAdi), is complex and depends among possible other factors on the pressure developed, lung volume and thoracic cage configuration (15, 38, 39), which may explain the poor or absent correlation between FT and Pdi in the studies discussed. Because chest cage configuration was not controlled in this study, we can however not derive any conclusions about the influence of the chest cage configuration on the relation between diaphragm thickness and effort.

The strong correlation between Pdi and strain as well as strain rate in the current study indicates that speckle tracking ultrasound derived parameters may provide good estimation of diaphragm effort, at least under inspiratory threshold loading. The performance of speckle tracking ultrasound under different loading conditions (isometric contractions, high inspiratory volume) remains to be evaluated.

**Future perspectives and clinical implications**

Regarding deformation analysis of the diaphragm, the software deriving the ST data has to be adapted to diaphragmatic ultrasonic morphology allowing quick, easy and reproducible analysis, preferably on-site. The offline setting of the actual data analysis is currently the only way to ensure proper data analysis, which hampers its use as bedside tool. In echocardiography most vendors provide a simplified software
tool for ST allowing measurements on site.

Only a few days of controlled mechanical ventilation is associated with atrophy of the diaphragm (40, 41). The reduction in diaphragm force, assessed by bilateral magnetic stimulation of the phrenic nerves, is approximately 30% in the first 5-6 days of invasive mechanical ventilation, indicating the rapid development of diaphragm weakness (42). In spite of the growing evidence that diaphragm weakness develops in critically ill patients and contributes to weaning failure and thus prolonged ventilation (42-44), respiratory muscle function is poorly monitored in these patients. Importantly, current state of the art techniques for monitoring, such as EAdi and Pdi, are invasive, not widely available and interpretation may be rather complex (7). An ideal assessment of diaphragm function must be available at bedside, fast and easy to acquire and allow standardized quantification. Guiding of ventilator weaning, assessing diaphragm contractile force during spontaneous breathing trials and/or pressure support ventilation might allow to adapt ventilation and weaning protocols individually. Both sufficient loading and prevention of excessive loading are decisive for weaning success. ST of the diaphragm might serve as diagnostic tool that can provide direct inside in diaphragm activity and force generation.

In conclusion, speckle tracking ultrasound as a non-invasive technique can be used to detect stepwise increases of diaphragmatic effort. Deformation (strain) and deformation velocity (strain rate) were highly correlated with transdiaphragmatic pressure and electric activity of the diaphragm. Speckle tracking ultrasound might serve as reliable tool to guide weaning at the bedside in the future.
References


Part II
Interventions
Automated patient-ventilator interaction analysis during non-invasive NAVA and pressure support ventilation in patients with COPD

Jonne Doorduin, Christer A. Sinderby, Jennifer Beck, Johannes G. van der Hoeven, Leo M.A. Heunks

Abstract

Introduction: Delivering synchronous assist during non-invasive ventilation (NIV) is challenging with flow or pressure controlled ventilators, especially in patients with chronic obstructive pulmonary disease (COPD). Neurally adjusted ventilatory assist (NAVA) uses diaphragm electrical activity (EAdi) to control the ventilator. We evaluated patient-ventilator interaction in patients with COPD during NIV with pressure support ventilation (PSV) and NAVA using an recently introduced automated analysis.

Methods: Twelve COPD patients underwent three 30-min trials: 1) PSV with dedicated NIV ventilator (NIV-PSV_Vision), 2) PSV with ICU ventilator (NIV-PSV_Servo-I), and 3) with NIV-NAVA. EAdi, flow, and airway pressure were recorded. Patient-ventilator interaction was evaluated by comparing airway pressure and EAdi waveforms with automated computer algorithms. The NeuroSync index was calculated as the percentage of timing errors between airway pressure and EAdi.

Results: The NeuroSync index was higher (larger error) for NIV-PSV_Vision (24 [15-30] %) and NIV-PSV_Servo-I (21 [15-26] %) compared to NIV-NAVA (5 [4-7] %; p <0.001). Wasted efforts, trigger delays and cycling-off errors were less with NAVA (p <0.05 for all). The NeuroSync index and the number of wasted efforts were strongly correlated ($r^2=0.84$), with a drastic increase in wasted efforts after timing errors reach 20%.

Conclusions: In COPD patients, non-invasive NAVA improves patient-ventilator interaction compared to PSV, delivered either by a dedicated or ICU ventilator. The automated analysis of patient-ventilator interaction allowed for an objective detection of patient-ventilator interaction during NIV. In addition, we found that progressive mismatch between neural effort and pneumatic timing is associated with wasted efforts.
Introduction

Non-invasive ventilation (NIV) plays an important role in managing patients with acute respiratory failure, in particular in patients with chronic obstructive pulmonary disease (COPD). In patients with acute hypercapnic exacerbation of COPD, NIV improves outcome (1-3). Accordingly, NIV utilization has increased over time among patients hospitalized for acute exacerbation of COPD, whereas the need for intubation has declined (2). Despite these positive reports, some patients treated with NIV fail and require invasive mechanical ventilation (3, 4). Poor patient-ventilator interaction may contribute to NIV failure (5, 6). Delivering synchronous non-invasive assist is challenging with flow or pressure controlled systems (7), especially when using excessively leaky or highly compliant interfaces (8). Using ventilators not dedicated to NIV, up to 46% of patients exhibit severe asynchrony (9). The introduction of dedicated NIV ventilators and NIV algorithms in ICU ventilators improved patient-ventilator interaction, yet their performance varies and the inherent limitations of using flow or pressure to control assist remain (10).

As recommended (11, 12), patient-ventilator interaction can be evaluated by using the diaphragm electrical activity (EAdi) (13). Besides its monitoring capabilities, EAdi is used during neurally adjusted ventilatory assist (NAVA) as a controller signal for ventilatory assist (14). Recent studies in heterogeneous groups of critically ill patients show that non-invasive NAVA (NIV-NAVA) improves patient-ventilator interaction relative to non-invasive pressure support ventilation (NIV-PSV) (15-18).

To our knowledge, there are no studies of patient-ventilator interaction strictly in COPD patients receiving NIV-NAVA, while these patients are more likely to exhibit severe patient ventilator asynchrony (19). In addition, no study has used the EAdi signal to evaluate patient-ventilator interaction with dedicated NIV ventilators. Moreover, a new automated analysis method has recently been introduced in this journal for quantifying patient-ventilator interaction in a standardized fashion (20). This automated analysis allows detection of asynchronies, such as wasted efforts, but also makes it easy to detect the more subtle dyssynchronies, such as trigger delays and cycling-off errors. The present study is the first to use this analysis method to quantify patient-ventilator interaction during non-invasive ventilation.

For the above-stated reasons, the aim of present study was to evaluate patient-ventilator interaction, using an automated analysis, in COPD patients with NIV-PSV delivered by a dedicated NIV ventilator, and NIV-PSV and NIV-NAVA delivered by an ICU ventilator.
Materials and methods

Study subjects
Adult patients with acute respiratory failure and a medical history of COPD, admitted to the ICU for non-invasive ventilation were eligible for inclusion in the study. Patients with a known neuromuscular disorder, severe hypoxemic failure (PaO₂/FiO₂ <100 mmHg), or hemodynamic instability requiring high dose norepinephrine (>0.5 µg/kg/min) were excluded. The study was approved by the ethical committee of the Radboud University Medical Center (NL33351.091.11) and is in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments. All patients gave their informed consent prior to the study.

Study design
All patients undergoing NIV in our hospital receive a nasogastric tube to allow adequate feeding and prevent gastric hyperinflation. COPD patients undergoing NIV receive a nasogastric tube with a multiple array of electrodes placed at the distal end (NAVA catheter, 12 French; Maquet Critical Care, Solna, Sweden). Correct positioning was established by use of dedicated software. After enrollment and clinical stabilization, each patient received three 30-min ventilation protocols in the following order:

1. Pressure support ventilation with the BiPAP Vision (Philips Respironics, Best, The Netherlands), a dedicated NIV ventilator, with pressure support and positive end expiratory pressure (PEEP) levels set by the treating physician (NIV-PSV<sub>Vision</sub>).
2. Pressure support ventilation with the Servo-I (Maquet Critical Care, Solna, Sweden, NIV software v3.0), an ICU ventilator with NIV algorithm, with similar PEEP and pressure support levels (NIV-PSV<sub>Servo</sub>).
3. NAVA with the Servo-I (Maquet Critical Care, Solna, Sweden, NIV software v3.0), where NAVA level was adjusted to match peak pressure of NIV-PSV, using manufacturer supplied software (NIV-NAVA).

BiPAP Vision uses the Auto-Trak Sensitivity algorithm to trigger and cycle off the ventilator and cannot be set individually. With NAVA, the back-up mode for triggering was set at flow triggering. In order to reduce the amount of leak on ventilator performance, we chose to use a tightly-fitted oro-nasal mask (Respironics PerforMax, Philips, Best, The Netherlands), a frequently used interface (21). Switching between ventilators required modifications in measurement setup and short disconnection of the patient from the ventilatory circuit. In order to minimize discontinuation of assist, the order of interventions were not randomized.
At the end of each ventilator mode, respiratory discomfort was scored by use of a Visual Analog Scale (from 0 mm [no discomfort] to 100 mm [maximal imaginable discomfort]) and arterial blood gas analysis was performed from an indwelling arterial line.

**Data acquisition**
Flow, airway pressure (Paw) and EAdi were acquired from the serial port of the Servo-I at a sampling rate of 62.5 Hz and recorded using dedicated acquisition software (Neurovent Research Inc., Toronto, Canada). The BiPAP Vision does not have a data output port. Therefore, flow was acquired by placing a pneumotachograph (Fleisch no. 3, Phipps & Bird, Virginia, USA) between the airflow port of the ventilator and its tubing, and Paw was acquired by placing a connection piece between the end of tubing (after the leakage port) and face mask, connected to a pressure transducer (range ± 50 kPa, Freescale, Tempe, AR). Both Paw and flow were recorded at a sampling rate of 62.5 Hz and synchronized with the EAdi using dedicated acquisition software (Neurovent Research Inc., Toronto, Canada).

**Data analysis**
Study parameters were calculated from a stable 5 minute period at the end of each mode on a breath-by-breath basis using a software routine developed for Matlab (Mathworks, Natick, MA). Measuring tidal volume by expiratory flow integration is not precise in the presence of leaks, therefore tidal volumes are not presented in the manuscript. Neural respiratory rate was calculated as the number of EAdi peaks/min.

Patient-ventilator interaction was evaluated by comparing Paw and EAdi waveforms with automated computer algorithms (20). Trigger and cycle-off error (i.e. dyssynchrony) were calculated as percentages of neural inspiratory and expiratory time periods, respectively. Events where EAdi and Paw were completely dissociated (i.e. asynchrony), such as wasted efforts, auto-triggering, multiple assist during EAdi peak (double triggering) and multiple EAdi peaks during assist, were assigned 100% error. To estimate the overall extent of asynchrony and dyssynchrony we calculated the NeuroSync index by averaging the percentage errors for all breaths.

**Statistical analysis**
The D’Agostino & Pearson test was used to test the normality of distribution. NIV-PSV Vision, NIV-PSV Servo-I and NIV-NAVA were compared using the Friedman test with Dunn’s post-hoc testing. Exponential regression analysis using an least squares fit was performed to test the relationship between the NeuroSync index and wasted efforts. A p value <0.05 was considered significant. Statistical analyses were performed with
Graphpad Prism 5 (Graphpad software, San Diego, CA, USA). Results are reported as median with interquartile ranges.

**Results**

Twelve patients (1 female / 11 male) were enrolled. One patient was excluded from the offline analysis due to an EAdi signal with a too low amplitude for automated patient-ventilator interaction analysis (20). Patient characteristics and ventilator settings are shown in Table 1 and 2, respectively. After study completion NIV failed in two patients and invasive ventilation was required. From these two patients, one deceased.

**Breathing pattern and respiratory drive**

Results for breathing pattern and respiratory drive are presented in Table 3. EAdi amplitude was higher with NIV-PSV\textsubscript{Servo-I} compared to NIV-PSV\textsubscript{Vision} (\(p < 0.05\)). Peak airway pressure and peak flow were higher with NIV-PSV\textsubscript{Vision} (\(p < 0.05\)) compared to NIV-NAVA and NIV-PSV\textsubscript{Servo-I}.

**Patient - ventilator interaction**

Figure 1 depicts median values for trigger delays and cycling-off error during each mode for all individual patients. NIV-NAVA showed lowest trigger delay compared to NIV-PSV\textsubscript{Vision} and NIV-PSV\textsubscript{Servo-I} (\(p < 0.0001\)). NIV-PSV\textsubscript{Vision} and NIV-PSV\textsubscript{Servo-I} had comparable trigger delays, but NIV-PSV\textsubscript{Servo-I} showed more early cycling-off (\(p < 0.05\)). In absolute values, NIV-PSV\textsubscript{Vision} (95 ± 22 ms) and NIV-PSV\textsubscript{Servo-I} (91 ± 19 ms) showed more cycling-off error compared to NIV-NAVA (12 ± 6 ms); \(p < 0.05\).

Patient-ventilator interaction, calculated with the NeuroSync index, was significantly higher (larger error) with NIV-PSV\textsubscript{Vision} (24 [IQR 15-30] %) and NIV-PSV\textsubscript{Servo-I} (21 [IQR 15-26] %) compared to NIV-NAVA (5 [IQR 4-7] %; \(p < 0.001\)).

Figure 2 depicts the correlation between the number of wasted efforts and the NeuroSync index. The relationship shows as timing errors progressively increased with NIV-PSV\textsubscript{Servo-I} and NIV-PSV\textsubscript{Vision} a positive association with the number of wasted efforts, which was certainly more pronounced above 20% error.

For all three modes of NIV studied, Figure 3 shows a plot of the relative timing-errors of triggering (Y-axis) versus the relative timing error for cycling-off (X-axis), for every breath, in all patients. Based on the data from Figure 2, we have inserted a box suggesting “acceptable” synchrony to be \(\leq 20\%\) of neural timings, whereas larger errors (> 20%) represent dyssynchrony.
In the form of a pie chart, Figure 4 plots the distribution of synchrony (<20% error, i.e. inside the box in Figure 3), dyssynchrony (>20% error, i.e. outside the box in Figure 3), and asynchronies for each mode. Wasted efforts were the most prevalent type of asynchrony and differed between ventilator modes ($p < 0.001$). Post-hoc analysis indicated significantly more wasted efforts with NIV-PSV$_{\text{Vision}}$ compared to NIV-NAVA. Other asynchronies, such as multiple EAdi during assist, double triggering and auto-triggering were uncommon.

**Blood gas values and respiratory discomfort**

There were no differences in blood gas values (Table 4) and respiratory discomfort with NIV-PSV$_{\text{Vision}}$ (45 [IQR 31-69] mm), NIV-PSV$_{\text{Servo-I}}$ (60 [IQR 41-65] mm), and NIV-NAVA (45 [IQR 33-75] mm).

**Table 1.** Patient characteristics at study inclusion

<table>
<thead>
<tr>
<th>#</th>
<th>Age (y)</th>
<th>BMI (kg/m$^2$)</th>
<th>FEV1 (% pred.)</th>
<th>FVC (% pred.)</th>
<th>FEV1 /FVC</th>
<th>GOLD class.</th>
<th>PF ratio</th>
<th>Reason for admission</th>
<th>Total NIV duration (days)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>37</td>
<td>25</td>
<td>316</td>
<td>I</td>
<td></td>
<td></td>
<td></td>
<td>haemoptysis</td>
<td>5</td>
</tr>
<tr>
<td>2</td>
<td>74</td>
<td>23</td>
<td>308</td>
<td>I</td>
<td></td>
<td></td>
<td></td>
<td>exacerbation COPD</td>
<td>3</td>
</tr>
<tr>
<td>3</td>
<td>68</td>
<td>38</td>
<td>42</td>
<td>III</td>
<td>185</td>
<td></td>
<td></td>
<td>exacerbation COPD</td>
<td>3</td>
</tr>
<tr>
<td>4</td>
<td>67</td>
<td>34</td>
<td>70</td>
<td>II</td>
<td>176</td>
<td></td>
<td></td>
<td>pneumonia</td>
<td>4</td>
</tr>
<tr>
<td>5</td>
<td>67</td>
<td>27</td>
<td>100</td>
<td>II</td>
<td>180</td>
<td></td>
<td></td>
<td>exacerbation COPD</td>
<td>5</td>
</tr>
<tr>
<td>6</td>
<td>64</td>
<td>26</td>
<td>43</td>
<td>III</td>
<td>220</td>
<td></td>
<td></td>
<td>trauma</td>
<td>3</td>
</tr>
<tr>
<td>7</td>
<td>58</td>
<td>26</td>
<td>143</td>
<td>II</td>
<td></td>
<td></td>
<td></td>
<td>exacerbation COPD</td>
<td>6</td>
</tr>
<tr>
<td>8</td>
<td>70</td>
<td>28</td>
<td>90</td>
<td>II</td>
<td>215</td>
<td></td>
<td></td>
<td>post-op lobectomy</td>
<td>2</td>
</tr>
<tr>
<td>9</td>
<td>78</td>
<td>22</td>
<td>64</td>
<td>II</td>
<td>110</td>
<td></td>
<td></td>
<td>exacerbation COPD</td>
<td>3</td>
</tr>
<tr>
<td>10</td>
<td>75</td>
<td>17</td>
<td>28</td>
<td>IV</td>
<td>219</td>
<td></td>
<td></td>
<td>exacerbation COPD</td>
<td>1</td>
</tr>
<tr>
<td>11</td>
<td>76</td>
<td>25</td>
<td>45</td>
<td>II</td>
<td>246</td>
<td></td>
<td></td>
<td>exacerbation COPD</td>
<td>5</td>
</tr>
</tbody>
</table>

Recent lung function tests for patient 1, 2 and 7 were unavailable in our hospital, but clinical picture of these patients were consistent with COPD and patient correspondence stated a history of COPD. BMI = body mass index; FVC = forced vital capacity; FEV1 = forced expired volume in 1 second; GOLD class. = Global Initiative for Chronic Obstructive Lung Disease classification; PF ratio = $\text{PaO}_2/\text{FiO}_2$.  

Patient-ventilator interaction during NIV in patients with COPD
### Table 2. Ventilator settings

<table>
<thead>
<tr>
<th>Patient</th>
<th>PS level (cmH₂O)</th>
<th>PS rise-time (s)</th>
<th>PS cycle-off (% peak flow)</th>
<th>NAVA level (cmH₂O/µV)</th>
<th>NAVA trigger (µV)</th>
<th>PEEP (cmH₂O)</th>
<th>FiO₂ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>6</td>
<td>0.20</td>
<td>30</td>
<td>0.1</td>
<td>0.5</td>
<td>6</td>
<td>70</td>
</tr>
<tr>
<td>2</td>
<td>10</td>
<td>0.20</td>
<td>50</td>
<td>0.8</td>
<td>0.5</td>
<td>8</td>
<td>50</td>
</tr>
<tr>
<td>3</td>
<td>8</td>
<td>0.20</td>
<td>70</td>
<td>0.4</td>
<td>0.5</td>
<td>8</td>
<td>50</td>
</tr>
<tr>
<td>4</td>
<td>5</td>
<td>0.20</td>
<td>50</td>
<td>0.1</td>
<td>1.0</td>
<td>7</td>
<td>60</td>
</tr>
<tr>
<td>5</td>
<td>6</td>
<td>0.20</td>
<td>50</td>
<td>0.1</td>
<td>0.5</td>
<td>4</td>
<td>40</td>
</tr>
<tr>
<td>6</td>
<td>5</td>
<td>0.05</td>
<td>50</td>
<td>5.0</td>
<td>0.5</td>
<td>5</td>
<td>30</td>
</tr>
<tr>
<td>7</td>
<td>10</td>
<td>0.00</td>
<td>50</td>
<td>0.2</td>
<td>0.5</td>
<td>6</td>
<td>55</td>
</tr>
<tr>
<td>8</td>
<td>6</td>
<td>0.20</td>
<td>50</td>
<td>0.2</td>
<td>0.5</td>
<td>6</td>
<td>45</td>
</tr>
<tr>
<td>9</td>
<td>6</td>
<td>0.20</td>
<td>50</td>
<td>0.2</td>
<td>0.5</td>
<td>6</td>
<td>70</td>
</tr>
<tr>
<td>10</td>
<td>8</td>
<td>0.00</td>
<td>60</td>
<td>0.1</td>
<td>0.5</td>
<td>5</td>
<td>35</td>
</tr>
<tr>
<td>11</td>
<td>6</td>
<td>0.20</td>
<td>50</td>
<td>0.2</td>
<td>0.5</td>
<td>6</td>
<td>40</td>
</tr>
</tbody>
</table>

Pressure support (PS) levels and rise time hold for both ventilators, whereas cycle-off criteria is only set for NIV-PSV₅₅. The BiPAP Vision uses the Auto-Trak Sensitivity algorithm to trigger and cycle off the ventilator and cannot be set individually. PEEP and FiO₂ were similar for all three ventilatory modes. FiO₂ = inspired oxygen fraction; NAVA = neurally adjusted ventilatory assist; PEEP = positive end-expiratory pressure.

### Table 3. Breathing pattern and respiratory drive

<table>
<thead>
<tr>
<th></th>
<th>NIV-PSVᵥision</th>
<th>NIV-PSV₅₅</th>
<th>NIV-NAVA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peak EAdi (µV)</td>
<td>25.6 (18.6 - 43.5)*</td>
<td>34.7 (18.8 – 49.0)</td>
<td>23.8 (17.1 - 48.0)</td>
</tr>
<tr>
<td>Peak airway pressure (cmH₂O)</td>
<td>15.3 (13.0 - 18.5)**</td>
<td>12.5 (10.4 - 15.2)</td>
<td>12.9 (11.7 - 16.0)</td>
</tr>
<tr>
<td>Inspiratory peak flow (L/min)</td>
<td>92.5 (72.1 - 110.0)**</td>
<td>54.1 (46.8 - 63.2)</td>
<td>45.6 (38.7 - 61.1)</td>
</tr>
<tr>
<td>Neural resp. rate (breaths/min)</td>
<td>22.7 (17.6 - 27.0)</td>
<td>25.2 (18.5 - 28.2)</td>
<td>25.1 (18.3 - 31.7)</td>
</tr>
</tbody>
</table>

EAdi = electrical activity of the diaphragm; NAVA = neurally adjusted ventilatory assist; NIV = non-invasive ventilation; PSV = pressure support ventilation. * NIV-PSVᵥision vs. NIV-PSV₅₅ (p < 0.05), ** NIV-PSVᵥision vs. NIV-NAVA (p < 0.05).
Patient-ventilator interaction during NIV in patients with COPD

Figure 1. Trigger delay (left) and cycling-off error (right) for the different ventilator modes. Y-axis for cycle-off error: positive values indicates late cycling-off, and negative values indicate early cycling-off. * p < 0.05. NAVA = neurally adjusted ventilatory assist; NIV = non-invasive ventilation; PSV = pressure support ventilation.

Figure 2. Correlation between the number of wasted efforts and the NeuroSync index. Note that for this regression analysis, the NeuroSync index was recalculated without wasted efforts to avoid mathematically coupled variables, and is thus consequently primarily a measure of dyssynchrony (trigger and cycle-off errors). Accordingly this correlation shows that progressive dyssynchrony, increases the number of wasted efforts. NAVA = neurally adjusted ventilatory assist; NIV = non-invasive ventilation; PSV = pressure support ventilation.
Figure 3. Breath density graph for relative trigger (Y-axis) and cycling-off (X-axis) errors, for all breaths in all patients, during each ventilator mode. The small white “box” in the center of each graph indicates the limit between synchrony (neural efforts matched to assist delivery with less than 20% error - inside the box) and dyssynchrony (neural efforts poorly related to assist delivery, > 20% error - outside the box). These breath-density graphs show for NIV-NAVA a concentrated breath density in the center, which should be anticipated since it is driven by EAdi. With NIV-PSV_{Vision} and NIV-PSV_{Servo-I} breaths are more spread-out and have considerable proportions of dyssynchronous breaths compared to NIV-NAVA. NAVA = neurally adjusted ventilatory assist; NIV = non-invasive ventilation; PSV = pressure support ventilation.

Figure 4. Percentage of synchronous, dyssynchronous and asynchronous (wasted efforts, auto-triggering, multiple EAdi during assist, and multiple assist during EAdi) breaths for the different ventilator modes. NAVA = neurally adjusted ventilatory assist; NIV = non-invasive ventilation; PSV = pressure support ventilation.
### Table 4. Blood gas values

<table>
<thead>
<tr>
<th></th>
<th>NIV-PSV Vision</th>
<th>NIV-PSV Servo</th>
<th>NIV-NAVA</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>7.38 (7.36 - 7.46)</td>
<td>7.38 (7.36 - 7.45)</td>
<td>7.38 (7.36 - 7.45)</td>
</tr>
<tr>
<td>PaO₂ (mmHg)</td>
<td>92 (77 - 106)</td>
<td>105 (84 - 113)</td>
<td>95 (77 - 98)</td>
</tr>
<tr>
<td>PaCO₂ (mmHg)</td>
<td>44 (39 - 64)</td>
<td>44 (33 - 59)</td>
<td>41 (32 - 60)</td>
</tr>
<tr>
<td>HCO₃⁻ (mmol/L)</td>
<td>27 (23 - 32)</td>
<td>26 (21 - 31)</td>
<td>27 (22 - 30)</td>
</tr>
</tbody>
</table>

NAVA = neurally adjusted ventilatory assist; NIV = non-invasive ventilation; PSV = pressure support ventilation.

### Discussion

This study provides insight into the interaction between patient and ventilator during noninvasive ventilation with different types of ventilators and modes in patients with COPD. First, we show that neurally adjusted non-invasive ventilation synchronizes assist to inspiratory effort in patients with COPD, whereas dedicated NIV ventilator or ICU ventilator pressure support modes do not ensure acceptable patient-ventilator interaction in individual patients. Second, wasted efforts increase drastically after timing errors between EAdi and airway pressure reach 20%. Third, automated analysis of patient-ventilator interaction using computer algorithms allows objective detection of patient-ventilator interaction during NIV.

### Patient-ventilator interaction

For effective unloading of the respiratory muscles with NIV, the ventilator should cycle in synchrony with the patient’s neural respiratory drive(5). Our results are consistent with previous studies that showed improved patient-ventilator interaction with neurally compared to pneumatically controlled mechanical ventilation(15-18), however several differences between these and the current study should be noted. First, we included only patients with COPD, which are more likely to exhibit poor patient-ventilator interaction (19). Second, dedicated NIV-NAVA and NIV-PSV software was used instead of software for invasive ventilation in the previous studies (16, 17). This is important as the software for invasive ventilation lacks leakage compensation thereby allowing auto-triggering at high leakage. Indeed, auto-triggering up to 6 breaths/min was found with NIV-NAVA using the invasive software (16), whereas we found only up to 1 breath/min. Third, a dedicated NIV ventilator was evaluated in the present study.
In bench-test comparisons, including the ventilator used in our study, PSV delivered by dedicated NIV ventilators allowed better patient-ventilator interaction than ICU ventilators with NIV algorithms (10, 22). Lastly, an automated analysis method for quantifying patient-ventilator asynchronies and the more subtle dyssynchronies was used (20), allowing a more objective detection of patient-ventilator interaction.

The present study showed a small trigger delay with NIV-NAVA, which substantially increased with NIV-PSV\textsubscript{Servo-I} and NIV-PSV\textsubscript{Vision}. These findings agrees with previous work comparing NIV-PSV\textsubscript{Servo-I} and NIV-NAVA (15), but was opposing previous bench-test showing longer trigger delay for NIV-PSV\textsubscript{Servo-I} compared to NIV-PSV\textsubscript{Vision} (22). NAVA triggers on the increase in EAdi and thus represents the duration to increase EAdi, to process the signal and to open the inspiratory valve. Our average trigger delay of about 50 ms with NIV-NAVA is in the range of previously reported for NIV-NAVA (15, 16, 18). In contrast, pneumatic triggering is more complex and considerably affected by leakage, which can only partly compensated for by dedicated NIV algorithms (7).

Synchronized termination of assist is another key component to maintain good patient-ventilator interaction. As depicted in Figure 1, NIV-PSV\textsubscript{Vision} showed large inter-subject variability in early and late cycling off, whereas NIV-PSV\textsubscript{Servo-I} showed primarily early cycling-off. Cycling-off error in NIV-NAVA was negligible, which could be anticipated since its definition for cycling-off is similar to the algorithm used to quantify cycling-off error (70\% of peak EAdi). These findings agree with previous suggestions that NIV-algorithms for ICU-ventilators tend to increase the incidence of premature cycling-off (7).

Wasted efforts are inspiratory efforts not rewarded by ventilatory assist, which can increase work of breathing (5, 6). In the present study, 4.3\% and 2.5\% of inspiratory efforts were unnoticed by the ventilator for NIV-PSV\textsubscript{Vision} and NIV-PSV\textsubscript{Servo-I} respectively. In contrast, NIV-NAVA effectively prevented wasted efforts, confirming previous studies (15, 16, 18). Furthermore, as depicted in Figure 2, we found that wasted efforts increase drastically after timing errors reach 20\%. This suggests that the limits of the NeuroSync index and the definition of “acceptable” synchrony should be kept below 20\%, as indicated by the centered boxes in Figure 3.

**Breathing pattern and respiratory drive**

EAdi in the present study was higher with NIV-PSV\textsubscript{Servo-I} compared to NIV-PSV\textsubscript{Vision}, which is difficult to explain. Lack of difference in blood-gases or respiratory rates contradict that increased EAdi with NIV-PSV was ventilation related. Premature cycling-off with NIV-PSV\textsubscript{Servo-I} could be a probable cause for increased EAdi, since this results in unassisted inspiration in the last part of inspiration. It should also be noted that the design of the respiratory circuit and assist delivery of the BiPAP Vision is fundamentally different.
from the Servo-I. For example, the BiPAP Vision system has a large intentional leakage. Consequently, from Ohm’s law it follows that higher flow is required to maintain the preset pressure level (Table 3). Higher flow might have resulted in higher CO₂ clearance in the interface and upper airways and a consequent reduction in dead space leading to reduced respiratory drive.

**Clinical implications**

Good patient-ventilator interaction is one of the key factors for clinical success of NIV, thus solving poor patient-ventilator interaction in COPD patients is of potential clinical value. In our study, we demonstrate that progressive mismatch between timing of the patient’s neural drive and the response of the ventilator is associated with increased number of wasted efforts. It is tempting to speculate that improving synchrony between patient neural effort and ventilator assist improves outcome in COPD patients, but it should be noted that our study is a short-term physiological study performed in a centre with extensive experience in NIV, both with PSV and NAVA. In addition, an limitation of the present study is the limited number of patients which hamper drawing generalized conclusions.

Differences in patient-ventilator interaction between ventilator modes did not affect blood gas values, in particular pH, and respiratory discomfort. In part, this results from the timing of study inclusion, after initial stabilization on NIV. At inclusion in the study, blood pH (around 7.38) already increased, making it more difficult to detect changes in pH and respiratory discomfort caused by different ventilator modes. In this context it should also be mentioned that NIV modes were not performed in a random order. Nevertheless, we performed our measurement after initial stabilization on NIV making it unlikely that the patients’ respiratory status was worse at the beginning of the study than at the end. Future studies, that randomize between NAVA and PSV at admission, are necessary to ascertain whether or not improved patient-ventilator interaction in the acute phase of NIV translates to better NIV outcomes.

**Conclusions**

Automated analysis of patient-ventilator interaction showed that non-invasive NAVA improves patient-ventilator interaction compared to PSV in COPD patients. Moreover, this is not different when PSV is delivered by a dedicated NIV ventilator. In addition, progressive mismatch between neural effort and pneumatic timing is strongly associated with the number of wasted efforts. Whether NAVA is more successful in correcting pH in patients with acute hypercapnic exacerbation of COPD should be addressed in future studies that randomize between NAVA and PSV at admission.
Acknowledgements

The authors thank Norman Comtois from the Department of Medicine, Division of Critical Care Medicine, St. Michael’s Hospital, University of Toronto for his technical assistance.
References


Patient-ventilator interaction during NIV in patients with COPD
Chapter 7

Assisted ventilation in patients with acute respiratory distress syndrome: lung-distending pressure and patient-ventilator interaction

Jonne Doorduin, Christer A. Sinderby, Jennifer Beck, Johannes G. van der Hoeven, Leo M.A. Heunks

*Anesthesiology. 2015 Jul; 123(1):181-90*
Abstract

Background: In ARDS patients, the use of assisted mechanical ventilation is subject of debate. Assisted ventilation has benefits over controlled ventilation, such as preserved diaphragm function and improved oxygenation. Therefore, higher level of ‘patient control’ of ventilator assist may be preferable in ARDS. However, assisted modes may also increase the risk of high tidal volumes and lung-distending pressures. The present study aims to quantify how differences in freedom to control the ventilator, affect lung protective ventilation, breathing patternvariability, and patient ventilator-interaction.

Methods: Twelve ARDS patients were ventilated in randomized order with assist pressure control ventilation (PCV), pressure support ventilation (PSV) and neurally adjusted ventilatory assist (NAVA). Transpulmonary pressure, tidal volume, diaphragm electrical activity and patient-ventilator interaction were measured. Respiratory variability was assessed using the coefficient of variation of tidal volume.

Results: During inspiration, transpulmonary pressure was slightly lower with NAVA (10.3±0.7, 11.2±0.7, and 9.4±0.7 cmH₂O for PCV, PSV, and NAVA; P<0.01). Tidal volume was similar between modes (6.6 [5.7-7.0], 6.4 [5.8-7.0] and 6.0 [5.6-7.3] ml/kg for PCV, PSV, and NAVA), but respiratory variability was higher with NAVA (8.0 [6.4-10.0], 7.1 [5.9-9.0], and 17.0 [12.0-36.1] % for PCV, PSV, and NAVA; P<0.001). Patient-ventilator interaction improved with NAVA (6 [5-8] % error) compared to PCV (29 [14-52] % error) and PSV (12 [9-27] % error); P<0.0001.

Conclusions: In patients with mild to moderate ARDS, increasing freedom to control the ventilator maintains lung protective ventilation in terms of tidal volume and lung-distending pressure, but improves patient-ventilator interaction and preserves respiratory variability.
Assisted ventilation in patients with ARDS

Introduction

In patients with acute respiratory distress syndrome (ARDS), mechanical ventilation with positive-end expiratory pressure (PEEP), limited plateau airway pressure and low tidal volume is widely accepted as the ventilation strategy of choice to limit ventilator induced lung injury (1, 2). The use of assisted mechanical ventilation in ARDS is a subject of debate (3-5). Among other beneficial effects, assisted ventilation preserves diaphragm function and better resembles natural respiratory variability when compared to controlled mechanical ventilation (3, 4). This may be important, as increased variation in breathing pattern improves oxygenation, lung mechanics and enhances tidal distribution to the dependent lung regions (6, 7). However, during assisted ventilation, spontaneous breathing contributes to the transpulmonary pressure, (Ptp) potentially increasing the risk of ventilator-induced lung injury if either is excessive (4).

Neurally adjusted ventilatory assist (NAVA) is a ventilator mode that uses diaphragm electrical activity (EAdi) to control timing and level of support during inspiration, thereby introducing inherent feedback loops between patient and ventilator (8). It has been shown in patients with acute respiratory failure that increasing levels of support with NAVA unloads the respiratory muscles, but limits pressure increases due to down-regulation of EAdi (9-11). This suggests that intrinsic lung-protective feedback mechanisms may limit tidal volume and lung distending pressure. Recently, it has been shown in an animal model that not only the magnitude, but also the duration of mechanical stress within the respiratory cycle (inspiratory to expiratory time ratio) determines the severity of lung injury (12). This emphasizes the importance of synchrony between neural and mechanical inspiration.

Overall, it is rational to investigate the effects of increased patient freedom to control the ventilator on respiratory mechanics in patients with ARDS. Up to now data related to these effects are scarce in patients with ARDS (10, 13). Therefore, the aim of the current study was to investigate the effects of three modes of assisted ventilation on lung distending pressures, tidal volume, breathing variability and patient-ventilator interaction in ARDS patients. The selected ventilator modes cover a wide spectrum of patient freedom to control ventilator assist: 1) pressure control ventilation (PCV): a pressure limited mode of both mandatory and triggered assist of fixed duration; 2) pressure support ventilation (PSV): a pressure limited mode with flow-trigger and variable cycling-off, allowing the patient more freedom to time assist delivery to inspiratory effort; and 3) NAVA: a mode where the patient regulates both timing and magnitude of assist via neural inspiratory effort. We reasoned that intrinsic lung-protective feedback mechanisms maintain lung-protective ventilation
when patient freedom to control the ventilator is increased. Accordingly, the first hypothesis of our study was that with NAVA (maximal ‘patient freedom’) tidal volume and lung-distending pressure remain within clinically acceptable limits. The second hypothesis was that breathing pattern variability and patient-ventilator interaction in ARDS patients improve with increased freedom to control the ventilator.

Materials and methods

Study design and population

Twelve adult patients, fulfilling the Berlin definition of ARDS (14), were studied in a physiological study on the intensive care unit of the Radboud university medical center. Exclusion criteria were hemodynamic instability, contra-indications to changing a naso-gastric tube (i.e. recent nasal bleeding, upper airway/esophageal pathology or surgery) and previously known neuromuscular disorders.

The protocol was approved by the institutional review board (CMO regio Arnhem-Nijmegen, NL31557.091.10) and is in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments. Patient surrogate decision makers gave informed consent before study inclusion.

Ventilator and esophageal catheter

Patients were ventilated with a Servo-I ventilator (Maquet Critical Care, Sölna, Sweden). EAdi and esophageal pressure (Pes) were obtained with a multi-electrode esophageal catheter with balloon (Neurovent Research Inc., Toronto, Canada), as described previously (15, 16). Care was taken with correct positioning and inflation of the balloon using the Baydur method (17). Additional details on catheter positioning are provided in the supplemental material.

Study protocol

Initially, patients were ventilated in PSV mode for 30 minutes to verify feasibility of assisted ventilation. Subsequently, in a cross-over design, PCV, PSV, and NAVA were randomly applied for 30 minutes each. PSV and PCV level were matched to target a Vt of 6 ml/kg predicted bodyweight. With PCV, mechanical respiratory rate was set just below patient’s respiratory rate to allow patient-triggered breaths. NAVA level (cmH2O/µV) was set by the use of a dedicated window on the Servo-I ventilator. Briefly, in either PSV or PCV, the actual airway pressure is presented with an overlay of the predicted airway pressure (as if the patient were in the NAVA mode). The NAVA level was adjusted manually to try to match the predicted peak pressure to the actual
peak airway pressure. PEEP was maintained constant throughout the study period and set according to the higher PEEP/lower FiO₂ arm of the ARDSnet consensus (18). Additional details on ventilator settings are provided in the supplemental material. At the end of each mode, arterial blood was sampled for analysis. Drugs, including sedatives and fluids, were unchanged during the entire study.

**Data acquisition**
Flow, Paw and EAdi were acquired (sampling rate 100Hz) from the serial port of the Servo-I and resampled to 2kHz. Pes was acquired (sampling rate 2kHz) by connecting the balloon to a pressure transducer (range ±50 kPa, Freescale, Tempe, AR, USA) and A/D converter (DT3004, Data Translation, Marlboro, MA, USA). All signals were acquired synchronously using dedicated software (Neurovent Research Inc., Toronto, Canada).

**Data analysis**
Data were analyzed offline. All variables were calculated from a stable 5-min period at the end of each mode on a breath-by-breath basis using a software routine developed for Matlab (Mathworks, Natick, MA). For each breath, we defined the following variables during the inspiratory phase (as shown in Figure 1):

- Tidal volume, as the integral of the inspiratory flow over time.
- Peak EAdi.
- Mean, peak and end-inflation airway pressure (Paw\textsubscript{mean}, Paw\textsubscript{peak}, Paw\textsubscript{end inflation}).
- Mean, peak inspiratory deflection and end-inflation esophageal pressure, as change from baseline (ΔPes\textsubscript{mean}, ΔPes\textsubscript{peak}, ΔPes\textsubscript{end inflation}). Baseline was defined as esophageal pressure at start of neural inspiration.
- Mean, peak and end-inflation transpulmonary pressure, as change from baseline (ΔPtp\textsubscript{peak}, ΔPtp\textsubscript{mean} and ΔPtp\textsubscript{end inflation}). Transpulmonary pressure was calculated as the difference between Paw and Pes. Baseline was defined as transpulmonary pressure at start of neural inspiration.
- Transpulmonary pressure-time product (PTP\textsubscript{tp}) was calculated as the integral of transpulmonary pressure over time. PTP\textsubscript{tp} is a measure of duration of mechanical stress within the respiratory cycle. To determine the effect of patient-ventilator dyssynchrony, this variable was also calculated during the following phases of inspiration: A) trigger delay; B) synchronous overlap period between neural and mechanical inspiration; and C) cycle-off error.
Neural respiratory rate was calculated as the number of EAdi peaks per minute and mechanical respiratory rate as the number of Paw peaks per minute. Breath-by-breath variability was assessed by calculating the coefficient of variation (CV; [standard deviation/mean] x 100) for Vt and EAdi.

Patient-ventilator interaction was evaluated by comparing Paw and EAdi waveforms with a recently validated automated computer algorithm that reports the timing error between Paw and EAdi (19, 20). Briefly, automatic detection of the start of neural inspiration (EAdi$_{ON}$) was obtained by detecting a 0.5 µV increase in EAdi. The end of neural inspiration (EAdi$_{OFF}$) was automatically detected by finding when the EAdi had decreased to 70% of its peak. The onset of pressure support (P$_{ON}$) was automatically detected by searching for an increase in Paw of >3 cmH$_2$O. The termination of pressure support (P$_{OFF}$) was automatically detected by searching for the decrease in Paw. See Figure 1 for an example of these timings. EAdi and Paw timings were used to calculate the NeuroSync index as following. The trigger error (P$_{ON}$ - EAdi$_{ON}$) and cycle-off error (P$_{OFF}$ - EAdi$_{OFF}$) were calculated as a percentage of the neural inspiratory detection period and neural expiratory detection period, respectively. Neural inspiratory detection periods were defined as segments from one detected EAdi$_{OFF}$ to the next EAdi$_{OFF}$. Neural expiratory detection periods were defined as segments from one EAdi$_{ON}$ to the next EAdi$_{ON}$. Thus, an early trigger error could range between -100% and 0% and a late trigger error could range between 0% and 100%. In the same fashion, an early cycle-off error could range between -100% and 0% and a late cycle-off error could range between 0% and 100%. Breaths with an absolute error of more than 33% where defined as dyssynchronous breaths, and breaths with an absolute error of less than 33% as synchronous breaths. Asynchronous breaths, defined as a complete dissociation between EAdi and Paw (wasted efforts, auto-triggering, double-triggering), were assigned 100% error. The NeuroSync index was then calculated by averaging the errors for all breaths per patient per mode.

**Statistical analysis**

To compare modes, one-way ANOVA for repeated measures was performed or the Friedman test as its non-parametric equivalent. Post-hoc analysis was performed with the Student-Newman-Keuls (SNK) test or Dunn’s test, as its non-parametric equivalent, to correct for multiple comparisons. The effect of patient-ventilator dyssynchrony on PTP$_{ip}$ was analyzed using two-way ANOVA for repeated measures with Bonferroni’s post-test. Linear regression analysis was performed to test associations. For all tests, a two-tailed $P<0.05$ was considered significant. Data are described as mean ± standard error for parametric data or median [interquartile
range] for nonparametric data. Statistical analyses were performed with Prism 5 (Graphpad software, San Diego, CA, USA).

**Figure 1.** Representative example of measurements of the main variables studied. The displayed breath is during pressure control ventilation. All variables were calculated on a breath-by-breath basis for each mode and averaged over a 5 min stable respiratory pattern. Vertical dotted lines represent neural and mechanical respiratory cycles. Arrows indicate the different parameters that are calculated from the waveforms. Tidal volume is calculated as the integral of inspiratory flow over time. Transpulmonary pressure versus time product ($\text{PTP}_{\text{tp}}$) is calculated as the integral of transpulmonary pressure ($\text{Ptp}$) over time. $\text{PTP}_{\text{tp}}$ was calculated during the following phases of inspiration: A) trigger delay; B) synchronous overlap period between neural and mechanical inspiration; and C) cycle-off error. $P_{\text{ON}} - \text{EAdi}_{\text{ON}} =$ trigger delay and $P_{\text{OFF}} - \text{EAdi}_{\text{OFF}} =$ cycle off error. Abbreviations: $\text{EAdi} =$ electrical activity of the diaphragm; $\text{Paw} =$ airway pressure; $\text{Pes} =$ esophageal pressure.
Results

Table 1 reports patient characteristics and Table 2 reports ventilator settings for PCV, PSV and NAVA during the study.

Respiratory variables
Tidal volumes were equal between modes (Figure 2A). One patient, having lowest pH of 7.18 revealed excessive Vt both in PSV and NAVA (#4, solid circles Figure 2A), but not in PCV. There was no difference between modes for neural and ventilator respiratory rates, however respiratory rates varied widely between subjects (Figure 2B). The coefficient of determination ($r^2$) between neural and ventilator respiratory rates was high with NAVA (0.99, $P<0.0001$), 0.70 ($P=0.0007$) with PCV, and 0.61 ($P=0.0026$) with PSV. Minute ventilation was equal between modes (Table 3). Peak EAdi ranged from near-zero to 30 mV between patients (Figure 2C). Post-hoc analysis did not reveal any significant differences between modes for EAdi. Mean Paw ($P_{aw\text{mean}}$) during inspiration was lower with NAVA compared to both PCV and PSV (Figure 2D). Changes in pleural pressure during inspiration are expressed by $\Delta P_{es\text{mean}}$ and ranged from positive to high negative values. Group mean values for $\Delta P_{es\text{mean}}$ were higher during PCV, i.e. less negative swing, compared to PSV and NAVA (Figure 2E). Lung distending pressure, expressed by $\Delta P_{tp\text{mean}}$, was highest during PSV, lower during PCV and lowest during NAVA (Figure 2F). Peak and end-inflation $\Delta P_{es}$ and $\Delta P_{tp}$ during inspiration were slightly lower during PCV compared to PSV and NAVA, whereas these values were equal for Paw (Table 3).

The CV of EAdi was equal between modes, whereas the CV of Vt was higher with NAVA (Table 3). Linear regression analysis between CVs of EAdi and Vt are presented in Figure 3, which shows no relationship with PCV, a poor relationship with PSV and a good relationship with NAVA.

There was a significant difference in PTP$_{tp}$ between modes (one-way ANOVA, $P = 0.0018$). Post hoc analysis showed that PTP$_{tp}$ was significantly lower ($P < 0.05$) with NAVA ($6.9 \pm 0.8$ cmH2O·s) compared to PSV ($8.3 \pm 0.7$ cmH2O·s) and PCV ($9.5 \pm 0.8$ cmH2O·s).
### Table 1. Patient characteristics at study inclusion.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age (y)</th>
<th>Sex</th>
<th>BMI (kg/m²)</th>
<th>RASS</th>
<th>Days on MV</th>
<th>P/F ratio (mmHg)</th>
<th>ARDS etiology</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>72</td>
<td>M</td>
<td>25</td>
<td>-3</td>
<td>13</td>
<td>242</td>
<td>Pneumonia</td>
</tr>
<tr>
<td>2</td>
<td>71</td>
<td>M</td>
<td>27</td>
<td>-5</td>
<td>4</td>
<td>146</td>
<td>Pneumonia</td>
</tr>
<tr>
<td>3</td>
<td>61</td>
<td>F</td>
<td>48</td>
<td>-1</td>
<td>1</td>
<td>116</td>
<td>Urosepsis</td>
</tr>
<tr>
<td>4</td>
<td>49</td>
<td>M</td>
<td>26</td>
<td>-1</td>
<td>21</td>
<td>75</td>
<td>Acute pancreatitis</td>
</tr>
<tr>
<td>5</td>
<td>64</td>
<td>M</td>
<td>23</td>
<td>-4</td>
<td>1</td>
<td>150</td>
<td>Pneumonia</td>
</tr>
<tr>
<td>6</td>
<td>76</td>
<td>M</td>
<td>32</td>
<td>0</td>
<td>4</td>
<td>108</td>
<td>Pneumonia</td>
</tr>
<tr>
<td>7</td>
<td>48</td>
<td>M</td>
<td>27</td>
<td>-4</td>
<td>7</td>
<td>143</td>
<td>Acute pancreatitis</td>
</tr>
<tr>
<td>8</td>
<td>71</td>
<td>F</td>
<td>32</td>
<td>-4</td>
<td>6</td>
<td>175</td>
<td>Acute pancreatitis</td>
</tr>
<tr>
<td>9</td>
<td>68</td>
<td>M</td>
<td>24</td>
<td>-4</td>
<td>32</td>
<td>177</td>
<td>Pneumonia and mediastinitis</td>
</tr>
<tr>
<td>10</td>
<td>78</td>
<td>M</td>
<td>18</td>
<td>-3</td>
<td>5</td>
<td>115</td>
<td>Pneumonia</td>
</tr>
<tr>
<td>11</td>
<td>45</td>
<td>M</td>
<td>24</td>
<td>-4</td>
<td>11</td>
<td>165</td>
<td>Pneumonia</td>
</tr>
<tr>
<td>12</td>
<td>66</td>
<td>M</td>
<td>23</td>
<td>-5</td>
<td>10</td>
<td>120</td>
<td>Pneumonia</td>
</tr>
</tbody>
</table>

Abbreviations: ARDS = acute respiratory distress syndrome; F = female; M = male; MV = mechanical ventilation; RASS = Richmond Agitation Sedation Scale.

### Table 2. Ventilator settings.

<table>
<thead>
<tr>
<th>Patient</th>
<th>PCV level (cmH₂O)</th>
<th>PSV level (cmH₂O)</th>
<th>NAVA level (cmH₂O/μV)</th>
<th>PEEP (cmH₂O)</th>
<th>FiO₂</th>
<th>Insp. time in PCV (s)</th>
<th>Cycle-off in PSV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>17</td>
<td>17</td>
<td>1.3</td>
<td>14</td>
<td>0.40</td>
<td>0.8</td>
<td>50</td>
</tr>
<tr>
<td>2</td>
<td>14</td>
<td>14</td>
<td>1.4</td>
<td>14</td>
<td>0.45</td>
<td>0.5</td>
<td>50</td>
</tr>
<tr>
<td>3</td>
<td>12</td>
<td>12</td>
<td>1.0</td>
<td>16</td>
<td>0.65</td>
<td>1.0</td>
<td>50</td>
</tr>
<tr>
<td>4</td>
<td>12</td>
<td>10</td>
<td>2.2</td>
<td>16</td>
<td>0.55</td>
<td>0.9</td>
<td>30</td>
</tr>
<tr>
<td>5</td>
<td>8</td>
<td>8</td>
<td>1.7</td>
<td>14</td>
<td>0.60</td>
<td>0.7</td>
<td>50</td>
</tr>
<tr>
<td>6</td>
<td>12</td>
<td>12</td>
<td>2.5</td>
<td>12</td>
<td>0.60</td>
<td>1.2</td>
<td>30</td>
</tr>
<tr>
<td>7</td>
<td>6</td>
<td>6</td>
<td>1.6</td>
<td>16</td>
<td>0.50</td>
<td>0.8</td>
<td>50</td>
</tr>
<tr>
<td>8</td>
<td>14</td>
<td>16</td>
<td>4.0</td>
<td>10</td>
<td>0.55</td>
<td>0.9</td>
<td>50</td>
</tr>
<tr>
<td>9</td>
<td>16</td>
<td>16</td>
<td>4.1</td>
<td>14</td>
<td>0.43</td>
<td>0.5</td>
<td>30</td>
</tr>
<tr>
<td>10</td>
<td>11</td>
<td>11</td>
<td>0.5</td>
<td>16</td>
<td>0.50</td>
<td>0.7</td>
<td>30</td>
</tr>
<tr>
<td>11</td>
<td>9</td>
<td>9</td>
<td>2.0</td>
<td>14</td>
<td>0.40</td>
<td>0.8</td>
<td>50</td>
</tr>
<tr>
<td>12</td>
<td>12</td>
<td>12</td>
<td>0.8</td>
<td>18</td>
<td>0.80</td>
<td>0.7</td>
<td>30</td>
</tr>
</tbody>
</table>

Abbreviations: I:E ratio = ratio between inspiratory and expiratory time; NAVA = neurally adjusted ventilatory assist; PCV = pressure control ventilation; PEEP = positive end-expiratory pressure; PSV = pressure support ventilation.
Figure 2. A) Tidal volume (Vt), B) ventilator and neural respiratory rate, C) electrical activity of the diaphragm (EAdi), D) mean airway pressure (Pawmean), E) mean esophageal pressure (ΔPesmean), and F) mean transpulmonary pressure (ΔPtpmean) for each ventilation mode. Filled circle symbol in panel A represents patient #4. Horizontal bars represent mean values (B, D-F) or median values (A, C). P-values in each panel represent result of one-way ANOVA for repeated measures (B, D-F) or Friedman test (A, C). Asterisks represent significant difference (P < 0.05) between indicated modes following post-hoc analysis with SNK’s test. Abbreviations: NAVA = neurally adjusted ventilatory assist; PCV = pressure control ventilation; PSV = pressure support ventilation.
Table 3. Breath parameters and variability.

<table>
<thead>
<tr>
<th></th>
<th>PCV</th>
<th>PSV</th>
<th>NAVA</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Minute Ventilation (L/min)</td>
<td>10.8 [7.2 - 13.2]</td>
<td>9.9 [8.7 - 12.9]</td>
<td>10.3 [9.0 - 12.6]</td>
<td>0.779</td>
</tr>
<tr>
<td>Pawpeak (cmH₂O)</td>
<td>28 ± 1</td>
<td>27 ± 1</td>
<td>29 ± 1</td>
<td>0.170</td>
</tr>
<tr>
<td>Pawend inflation (cmH₂O)</td>
<td>24 ± 1</td>
<td>25 ± 1</td>
<td>25 ± 1</td>
<td>0.684</td>
</tr>
<tr>
<td>∆Pespeak (cmH₂O)</td>
<td>-5 ± 1</td>
<td>-6 ± 1</td>
<td>-7 ± 1</td>
<td>0.027</td>
</tr>
<tr>
<td>∆Pesend inflation (cmH₂O)</td>
<td>3 [2 - 4]</td>
<td>1 [-5 - 2]</td>
<td>0 [-2 - 2]</td>
<td>0.039</td>
</tr>
<tr>
<td>∆Ptpppeak (cmH₂O)</td>
<td>16 ± 1</td>
<td>17 ± 1</td>
<td>17 ± 1</td>
<td>0.016</td>
</tr>
<tr>
<td>∆Ptpend inflation (cmH₂O)</td>
<td>8 ± 1 a, b</td>
<td>11 ± 1</td>
<td>11 ± 1</td>
<td>0.014</td>
</tr>
<tr>
<td>CV of Vt (%)</td>
<td>8.0 [6.4 - 10.0] b</td>
<td>7.1 [5.9 - 9.0] c</td>
<td>17.0 [12.0 - 36.1]</td>
<td>0.0005</td>
</tr>
<tr>
<td>CV of EAdi (%)</td>
<td>27.7 [22.5 - 40.5]</td>
<td>28.2 [23.8 - 38.6]</td>
<td>29.3 [22.7 - 39.5]</td>
<td>0.920</td>
</tr>
</tbody>
</table>

Data are described as mean ± standard error for parametric or median [interquartile range] for non-parametric data. P-values represent result of one-way ANOVA for repeated measures for parametric data or the Friedman test as its non-parametric equivalent. Annotations represent significant difference (P < 0.05) between indicated modes following post-hoc analysis with SNK’s (parametric data) or Dunn’s (non-parametric data) test, where a = PCV vs. PSV, b = PCV vs. NAVA, and c = PSV vs. NAVA. Abbreviations: CV = coefficient of variation; EAdi = electrical activity of the diaphragm; NAVA = neurally adjusted ventilatory assist; Pawpeak = peak airway pressure; PCV = pressure control ventilation; Pespeak = peak esophageal pressure; PSV = pressure support ventilation; Ptpppeak = delta peak transpulmonary pressure; Vt = tidal volume.

Figure 3. Linear regression analysis between coefficient of variation of electrical activity of the diaphragm (CV of EAdi) and coefficient of variation of tidal volume (CV of Vt) for each ventilation mode. Abbreviations: NAVA = neurally adjusted ventilatory assist; PCV = pressure control ventilation; PSV = pressure support ventilation.
Patient-ventilator interaction

The percentage of synchronous breaths was higher and trigger delay shorter with NAVA compared to PCV and PSV (Table 4). Cycle-off error was higher with PCV compared to PSV and NAVA (Table 4). Wasted efforts and double triggering were overall uncommon and equal between modes, whereas the rate of auto-triggering was different between modes (Table 4). The latter is explained by the fact that during PCV, 87.6 ± 16.8 % of the breaths were patient-triggered and 12.4 ± 16.8 % were ventilator controlled. The NeuroSync index, depicted in Figure 4A (0% error = perfect patient-ventilator interaction and 100% error = zero patient-ventilator interaction), indicated more than 20% error in seven patients during PCV, which improved during PSV and was below 20 % in all patients during NAVA. The higher cycle-off error with PCV resulted in an increased PTPtrp, as depicted in Figure 4B.

Blood gases

Table 5 shows arterial blood gas values at the end of each 30 minute interval. PaO₂ and PaCO₂ were significantly higher with NAVA compared to PCV (Table 5).

<table>
<thead>
<tr>
<th>Table 4. Effect of different ventilation modes on patient-ventilator interaction.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
</tr>
<tr>
<td>------------------</td>
</tr>
<tr>
<td>Synchrony (breaths)</td>
</tr>
<tr>
<td>Dyssynchrony (breaths)</td>
</tr>
<tr>
<td>Trigger delay (ms)</td>
</tr>
<tr>
<td>Cycle off error (ms)</td>
</tr>
<tr>
<td>Wasted efforts (breaths)</td>
</tr>
<tr>
<td>Auto-triggering (breaths)</td>
</tr>
<tr>
<td>Double triggering (breaths)</td>
</tr>
</tbody>
</table>

Data are described as mean ± standard error for parametric or median [interquartile range] for non-parametric data. \(P\)-value represents result of one-way ANOVA for repeated measures for parametric data or the Friedman test as its non-parametric equivalent. Annotations represent significant difference (\(P < 0.05\)) between indicated modes following post-hoc analysis with SNK’s (parametric data) or Dunn’s (non-parametric data) test, where \(^a\) = PCV vs. PSV, \(^b\) = PCV vs. NAVA, and \(^c\) = PSV vs. NAVA. Abbreviations: \(\text{EAdi} = \) electrical activity of the diaphragm; NAVA = neurally adjusted ventilatory assist; PCV = pressure control ventilation; PSV = pressure support ventilation. Values are given as median and interquartile range.
Assisted ventilation in patients with ARDS

Figure 4. A) NeuroSync index (19) of each ventilation mode. The NeuroSync index is an overall indicator of patient-ventilator interaction, where 0% error = perfect and 100% error = zero patient-ventilator interaction. Horizontal bar represent median value and P-value in panel represent result of Friedman test. Asterisk represents significant difference (P < 0.05) between indicated modes following post-hoc analysis with Dunn’s test. B) Transpulmonary pressure-time product (PTPtp) during different phases of patient-ventilation interaction. Bars represent mean ± standard error. P-value in panel represent result of two-way ANOVA for repeated measures. Asterisk represents significant difference (P < 0.05) between indicated modes following post-hoc analysis with Bonferroni’s test. Abbreviations: NAVA = neurally adjusted ventilatory assist; PCV = pressure control ventilation; PSV = pressure support ventilation.

Table 5. Effect of different ventilation modes on blood gas values

<table>
<thead>
<tr>
<th></th>
<th>PCV</th>
<th>PSV</th>
<th>NAVA</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>PaO2 (mmHg)</td>
<td>78 [68 - 84]</td>
<td>80 [69 - 97]</td>
<td>81 [69 - 91]</td>
<td>0.022</td>
</tr>
<tr>
<td>PaO2/FiO2 (mmHg)</td>
<td>153 ± 12</td>
<td>163 ± 16</td>
<td>163 ± 13</td>
<td>0.341</td>
</tr>
<tr>
<td>PaCO2 (mmHg)</td>
<td>46 ± 3</td>
<td>48 ± 3</td>
<td>49 ± 3</td>
<td>0.008</td>
</tr>
<tr>
<td>HCO3⁻ (mmol/L)</td>
<td>27.4 ± 2.1</td>
<td>27.5 ± 2.0</td>
<td>27.8 ± 2.0</td>
<td>0.235</td>
</tr>
</tbody>
</table>

Data are described as mean ± standard error for parametric or median [interquartile range] for non-parametric data. P-value represents result of one-way ANOVA for repeated measures for parametric data or the Friedman test as its non-parametric equivalent. Asterisks represent significant difference (P < 0.05) between PCV and NAVA following post-hoc analysis with SNK’s (parametric data) or Dunn’s (non-parametric data) test. Abbreviations: NAVA = neurally adjusted ventilatory assist; PCV = pressure control ventilation; PSV = pressure support ventilation. Values are given as mean ± standard error. a PCV vs. NAVA (P < 0.05).
Discussion

The present study reports the effects of different modes of assisted ventilation on lung-distending pressure, tidal volume, breathing pattern variability, and patient-ventilator interaction in ARDS patients. The three selected modes allow for different degrees of patient freedom to control the ventilator. The main findings of this study were that with increasing freedom to control the ventilator (1) tidal volume remains within the limits of lung-protective ventilation; and (2) lung distending pressures remain similar between modes. Even with NAVA, when the patient has a high degree of freedom to control the amount of support, tidal volume and lung distending pressures were similar to assist control ventilation, whereas breathing pattern variability and patient-ventilator interaction improved compared to PCV and PSV. Furthermore, prolonged inspiratory time during PCV resulted in a longer duration of mechanical stress.

Tidal volume and lung distending pressures

In the present study, Vt could be considered lung-protective in the majority of patients and was not affected by ventilator mode. Peak and end-inflation airway pressures (Table 3) were equal between modes, whereas mean airway pressures were lower during NAVA than during PCV and PSV (Figure 2D). Lower mean ventilator pressures with NAVA could be explained by the slower rate of pressurization with NAVA compared to a squared pressure waveform with PSV and PCV. These lower mean airway pressures do not necessary reflect a too low level of assistance during NAVA. As evidenced by the similar levels of EAdi with the three modes (Fig 2C), and the similar level of mean esophageal pressure compared to PSV. The lower level of mean esophageal pressure during PCV mainly reflects a too long inspiratory time during PCV. Although there were statistical differences in $\Delta Ptp_{\text{mean}}$, $\Delta Ptp_{\text{end inflation}}$, and $\Delta Ptp_{\text{peak}}$ between modes, these were small in absolute values. Based on a post-hoc analysis of the low tidal volume ARDSnet study (1), Hager and colleagues concluded that no safe plateau pressure exists (21). It can be derived from the first figure in their manuscript that reduction in plateau pressure from 30 cmH$_2$O to 25 cmH$_2$O is associated with reduction in mortality of $\sim$4%. In our study, differences in airway and transpulmonary pressures among modes were $\leq$3 cmH$_2$O and therefore probably of limited clinical significance.

Protection against excessive Ptp and Vt is likely mediated by vagal afferents sensitive to lung distension (22, 23). Indeed, our data demonstrate similar Vt and lung-distending pressures during PCV, PSV (both delivering fixed pressure) and NAVA (proportional control of pressure). Prevention of high Vt and Ptp may be mediated by
the Hering-Breuer inspiratory inhibiting (vagal) reflex where lung inflation at critical volume (i.e. lung stretch) terminates inspiration (24). However, in the present study in one patient a tidal volume >9 mL/kg was observed during PSV and NAVA. This patient had the lowest blood pH (7.18), consistent with the view that acid-base homeostasis is the primary goal of the respiratory centers in the brain stem and may be achieved at the expense of high Vt or Ptp (25). Based on this observation we suggest to be very cautious with the use of assisted breathing modes in patients with a very low pH.

**Breathing pattern variability**

Preservation of natural breathing variability and complexity during mechanical ventilation is considered important, as it may improve oxygenation, lung mechanics and gas distribution to the more dependent regions (6, 7). Breath-by-breath variability of Vt with NAVA was higher in comparison to PCV and PSV, whereas EAdi variability was similar. Accordingly, the ventilator output responded to the patient’s neural breathing effort during NAVA and not during pressure targeted assist (Figure 3), which is in accordance with previous findings (9, 26). By definition, a larger respiratory variability with NAVA increases the incidence of breaths with a high Vt (9). It is unknown whether this periodic delivery of breaths with a higher Vt during assisted mechanical ventilation is harmful. It could also be regarded as a type of sigh or recruitment maneuver, which have been shown to have moderate success for improving lung mechanics and gas exchange (27-29). In the present study oxygenation improved slightly with NAVA compared to PCV, but not compared to PSV. This might be explained by better lung recruitment of the dependent regions with NAVA (6). PaCO₂ was slightly lower during PCV compared to NAVA. This might have resulted from small differences in tidal volume, respiratory rate, and tidal volume distribution, nevertheless blood pH was unaffected.

**Patient-ventilator interaction**

With NAVA patient-ventilator interaction was with minimal error, whereas prevalence of patients with poor patient-ventilator interaction increased during PSV and PCV. This is consistent with recent findings in ARDS patients (10, 13) and in other patients with acute respiratory failure (30, 31). This is of potential clinical relevance, as patient-ventilator asynchrony has been associated with adverse clinical outcome, including prolonged mechanical ventilation (32, 33). For example, reversed triggering or breath-stacking in assist-control mode may contributing to ventilator induced lung injury (34, 35). See supplemental Figure 1 for an example of breath-stacking caused by reversed triggering in the current study. Experimental animal data suggests that
not only the magnitude but also the inspiratory cycle duration of lung distending pressure during PCV is associated with lung injury of increased severity (12, 36). In the present study, late cycling-off resulted in prolonged and unnecessary lung distending pressures with PCV, and to a lesser extent with PSV (Figure 4B). Thus, our results support the notion that poor patient-ventilator interaction and fixed duration of assist during spontaneous breathing is less lung protective than synchronized assist delivery in proportion to neural inspiratory effort.

**Clinical implications**

The role of assisted ventilation in ARDS has been extensively debated in recent years (4). The most striking evidence in opposition to assisted ventilation is the effect of muscle paralysis in the first 48 hours in ARDS patients with PaO\(_2\)/FiO\(_2\) <150 mmHg, which has been associated with improved outcome (37). Our findings may have implications for mechanical ventilation in mild to moderate ARDS following these first 48 hrs. First, our results suggest that it is reasonable from a physiological point of view to use an assisted ventilation mode with good patient-ventilator interaction without increasing the risk of excessive tidal volume and lung-distending pressure. Our data are supported by recent findings in a pig model of ARDS, that higher levels of spontaneous breathing reduce global stress and strain (38). Second, in terms of the patient’s inspiratory drive and effort, the present study revealed a high variability in EAdi and Pes values, ranging from over-assist to strenuous efforts (Figure 1). Also, a great dispersion in neural respiratory rate implies a large variety in respiratory drive in ARDS patients. These findings suggest that adjusting ventilator assist to target low tidal volumes resulted in different levels of neural drive in each study patient, probably due to a complex interaction between sedation, ventilatory mode and level of assist. Accordingly, monitoring Vt in conjunction with EAdi and/or Pes can aid to optimize ventilation in ARDS to prevent over-assist or insufficient assist and improve patient-ventilator interaction (39). The capabilities of EAdi for monitoring purposes were recently well demonstrated in trial to monitor dynamic intrinsic PEEP (40).

**Critique of the method**

Several limitations of the present study should be addressed. First, esophageal pressure was used as an estimate of pleural pressure. The validity of esophageal pressure for this purpose has been discussed before in the literature (41, 42). Second, the real lung distending pressure is transpulmonary pressure obtained under static conditions (zero-flow) to eliminate the resistive component in the equation of motion. Achieving and verifying static conditions in spontaneous breathing under assisted ventilation in critically ill patients is very cumbersome. Therefore
our measurements of lung distending pressures were performed during dynamic conditions, and cannot directly be compared to static transpulmonary pressures. As peak pressure is highly influenced by airway resistance and is not a reliable estimate of lung distending pressure, mean transpulmonary pressure during inspiration may be a better estimate under dynamic condition. Lastly, it should be recognized that our study is a single-center, physiological study, conducted in a heterogeneous group of ARDS patients with a wide range of sedation scores. Different conditions leading to ARDS, such as pulmonary versus extrapulmonary, might influence the effect on lung-protective ventilation while changing ventilator modes, when studied in large subgroups. In our study, each mode was studied for a period of 30 minutes. This allows for physiological comparisons and improves our understanding in the effects of these different modes on respiratory mechanics, patient-ventilator interaction and gas exchange. Large clinical trials that evaluate these different ventilator modes for several days in large groups of patients are needed to determine whether one mode is superior in outcome compared to the other modes.

In conclusion, in patients with mild to moderate ARDS, increasing patient freedom to control the ventilator maintains lung protective ventilation in terms of tidal volume and lung-distending pressure, but improves patient-ventilator interaction and preserves respiratory variability.

Acknowledgments

The authors thank Norman Comtois from the Department of Critical Care Medicine, St. Michael’s Hospital, Toronto, Ontario, Canada, for his technical assistance.
References


Supplemental material

Supplemental materials and methods

Esophageal catheter positioning and balloon dimensions
Correct positioning of the catheter was obtained using standard software supplied with the ventilator by the manufacturer, which results in centering of the esophageal balloon about 12 cm proximal to the diaphragm. Proper esophageal balloon position was confirmed by the presence of proportionality between deflections in airway pressure (Paw) and Pes during inspiratory efforts against an occlusion (1).

Detailed ventilator settings
Trigger sensitivity was set at 50% change of bias flow (2 L/min) and the inspiration/expiration ratio (I:E ratio) was set to obtain optimal patient-ventilator interaction based upon visual inspection of ventilator pressure and flow tracings. With PSV, trigger sensitivity and rise time were similar as in PCV. Inspiratory cycle off was adjusted for each patient based on flow and pressure waveforms to obtain a level where premature and late cycling-off was visually minimal. With NAVA, trigger sensitivity was 0.5 µV above baseline and cycle-off was manufacturer set at 70% of peak EAdi.

Supplemental References
Supplemental Figure 1. Breath-stacking caused by reversed triggering in patient #4 during pressure control ventilation. Note the increase in tidal volume during each second patient-triggered breath. EAdi = electrical activity of the diaphragm; Paw = airway pressure; Vt = tidal volume.
Chapter 8

Partial neuromuscular blocking during partial ventilator support in sedated patients with high tidal volumes

Jonne Doorduin, Joeke L Nollet, Lisanne H Roesthuis, Hieronymus W.H. van Hees, Laurent J. Brochard, Christer A. Sinderby, Johannes G. van der Hoeven, Leo M.A. Heunks

Abstract

Rationale: Controlled mechanical ventilation is used to deliver lung-protective ventilation in patients with acute respiratory distress syndrome. Despite recognized benefits, such as preserved diaphragm activity, partial support ventilation modes may be incompatible with lung-protective ventilation due to high tidal volume and high transpulmonary pressure. As an alternative to high dose sedatives and controlled mechanical ventilation, pharmacologically induced neuromechanical uncoupling of the diaphragm should facilitate lung-protective ventilation under partial support modes.

Objectives: To investigate whether partial neuromuscular blockade can facilitate lung-protective ventilation while maintaining diaphragm activity under partial ventilatory support.

Methods: In a proof-of-concept study, we enrolled ten patients with lung injury and a tidal volume >8 ml/kg under pressure support ventilation (PSV) and under sedation. After baseline measurements, rocuronium administration was titrated to target tidal volume of 6 ml/kg during neurally adjusted ventilatory assist (NAVA). Thereafter patients were ventilated in PSV and NAVA under continuous rocuronium infusion for 2 hours. Respiratory parameters, hemodynamic parameters and blood gas values were measured.

Measurements and Main Results: Rocuronium titration resulted in significant declines of tidal volume (9.3±0.6 to 5.6±0.2 ml/kg; \(p<0.0001\)), transpulmonary pressure (26.7±2.5 to 10.7±1.2 cmH\(_2\)O; \(p<0.0001\)) and diaphragm electrical activity (17.4±2.3 to 4.5±0.7 µV; \(p<0.0001\)) and could be maintained under continuous rocuronium infusion. During titration pH (7.42±0.02 to 7.35±0.02; \(p<0.0001\)) decreased, and mean arterial blood pressure (84±6 to 99±6 mmHg; \(p=0.0004\)) and heart rate (83±7 to 93±8 beats/min; \(p=0.0004\)) increased.

Conclusions: Partial neuromuscular blockade facilitates lung-protective ventilation during partial ventilatory support while maintaining diaphragm activity in sedated patients with lung injury.
Partial neuromuscular blocking in patients with high tidal volumes

Introduction

Mechanical ventilation is the cornerstone of treatment of patients presenting with acute respiratory distress syndrome (ARDS). Despite being lifesaving, mechanical ventilation can aggravate lung injury (1-4). A lung-protective ventilation strategy targeting low tidal volume ($V_T$) and limited plateau airway pressure under controlled mechanical ventilation early in the course of ICU admission has been shown to improve survival in ARDS patients (5, 6). However, the use of controlled modes generally requires high levels of sedation (7, 8), which is associated with adverse outcomes (9, 10). In particular, controlled mechanical ventilation is associated with the development of diaphragm dysfunction (11-13). For instance, in a landmark study Levine and colleagues demonstrated that only a few days of controlled mechanical ventilation results in diaphragm muscle fiber atrophy (11). In accordance with these data, Jaber and colleagues reported a marked reduction of in vivo diaphragmatic force in patients on mechanical ventilation (13). Recently, the frequent occurrence of diaphragm atrophy was also demonstrated in the ICU (14).

Modes of mechanical ventilation that allow spontaneous breathing, i.e. partial support ventilation, have recognized benefits including lung recruitment (15-17) and reduced risk of ventilator-induced diaphragm dysfunction (VIDD) (18-20). Recently, we demonstrated that in selected patients with mild to moderate ARDS partial support modes can deliver support within the limits of lung-protective ventilation (21). However, there are also patients in whom the transition from controlled modes to partial support modes is difficult because control of lung protection is lost. In partial support modes, these patients will generate high $V_T$, high transpulmonary pressures ($P_L$) and develop excessive work of breathing (WOB). Accordingly, patient selection and in particular timing for initiation of partial support ventilation is still under debate (22-24). In patients generating high $V_T$ despite low level of assist clinicians may prefer controlled ventilation, including high dose sedatives. As an alternative strategy, pharmacologically induced partial neuromechanical uncoupling of the respiratory muscles may facilitate a lung-protective ventilation strategy, while maintaining diaphragm activity. This may allow a safe compromise between the benefits and risks of spontaneous breathing during mechanical ventilation (25). Interestingly, Campbell (26) demonstrated that low dose curare, a neuromuscular blocking agent (NMBA) of the acetylcholine receptor, reduces maximum inspiratory pressure, while maintaining spontaneous breathing in awake healthy subjects. We have explored here the possibility to reach irreconcilable targets with partial ventilatory assist. The targets are on the one hand keeping the benefit of spontaneous breathing in patients with lung injury, which have been demonstrated quite strongly and include
a potential prevention against ventilator-induced diaphragm dysfunction, and on the other hand avoid the risk of excessive tidal volumes reached by high respiratory drive under partial ventilatory assist.

Accordingly, in the current study we investigated whether low dose neuromuscular blockers can facilitate a lung-protective ventilation strategy without loss of diaphragm activity during partial ventilatory support in lung injury patients with high tidal volumes. We hypothesized that partial neuromuscular blockade of the diaphragm allows the control of $V_t$ and $P_l$ to traditional lung-protective values, while maintaining diaphragm activity.

**Methods**

**Study design and population**
This proof-of-concept study was performed at the ICU of a university hospital. Adult patients admitted for ARDS were eligible if they were able to pneumatically trigger the ventilator and developed high $V_t$ (>8 ml/kg predicted bodyweight) during pressure support ventilation (PSV) of 12 cmH$_2$O. Exclusion criteria were recent use of NMBAs (<3 hours), pain or agitation (RASS > 0), metabolic acidosis, hemodynamic instability (i.e. high dose vasopressors or inotropes), elevated intracranial pressure, open chest/abdomen and past medical history of neuromuscular disorders.

The study was approved by the local ethics review committee and conducted in accordance with the Declaration of Helsinki. Patient surrogate decision makers gave written informed consent before study inclusion.

**Study protocol**
All patients were ventilated with the Servo-i ventilator (Maquet Critical Care, Sölna, Sweden). The study protocol consisted of three phases. Phase 1: baseline measurements, phase 2: titration of NMBA and phase 3: continuous infusion of NMBA (Figure 1). In phase 1, the physiological effects of different partial support modes were evaluated: assist-volume control ventilation (VCV) of 6 ml/kg, PSV of 12 cmH$_2$O (PSV-12), PSV of 6 cmH$_2$O (PSV-6) and neurally adjusted ventilatory assist (NAVA), where assist was titrated to equal $V_t$ as with PSV-12. Two levels of PSV were selected, where 12 cmH$_2$O reflects a reasonable level of support in these patients and 6 cmH$_2$O was selected to confirm high $V_t$ under low level of assist. In phase 2 (titration), rocuronium bromide (Fresenius Kabi, Zeist, The Netherlands) was titrated to obtain $V_t$ <=6 ml/kg (details in the supplemental material). Titration was deliberately performed in NAVA, as in this mode diaphragm activity and support are
Partial neuromuscular blocking in patients with high tidal volumes

tightly linked and thus preservation of diaphragm activity is guaranteed. In phase 3 (continuous infusion), patients were ventilated with PSV and NAVA for 1 hour per mode to evaluate feasibility of continuous partial neuromuscular blockade. PSV was chosen in addition to NAVA, as the former is the most widely used partial support mode. During phase 3 PSV level was set to obtain equal tidal volume as with NAVA. Positive end-expiratory pressure (PEEP) was set according clinical protocol and not changed. The sedation protocol is provided in the supplemental material.

**Figure 1.** Study protocol. Phase 1: different modes of partial support ventilation were evaluated without patients receiving neuromuscular blockade. NAVA level (cmH₂O/µV) was titrated to equal support under PSV-12. Phase 2: following every bolus rocuronium an average VT of 10 breaths was calculated to evaluate whether the target of 6 ml/kg was reached. Phase 3: PSV and NAVA were applied in random order for 1 hour with continuous rocuronium infusion. PSV level was set to obtain equal support as with NAVA. If required an additional bolus rocuronium was given to control VT. NAVA = neurally adjusted ventilatory assist; PSV = pressure support ventilation; VCV = volume control ventilation; VT = tidal volume.

**Data acquisition and analysis**
Flow, ventilatory pressure (Pvent), electrical activity of the diaphragm (EAdi), esophageal pressure (Pes), gastric pressure (Pga), blood pressure, heart rate and arterial oxygen saturation (SaO₂) were acquired. Data acquisition and analysis details are provide in the supplemental material. The duration of phase 2 (titration) differs per patient, therefore data were analyzed at seven points: the first and last minute.
and five 1-min periods taken at equal time intervals in between. Blood samples were taken before phase 1 (baseline) and 2, and after phase 2 and 3.

**Statistical analysis**

To compare between ventilator modes and/or points in time, repeated measures ANOVA was performed (Prism 5, Graphpad, San Diego, CA, USA). If appropriate, post-hoc analysis was performed. A two-tailed \( p < 0.05 \) was considered significant. Data are described as mean ± SEM, except as stated otherwise.

**Results**

All patients fulfilled the Berlin criteria for ARDS at ICU admission with a \( \text{PaO}_2/\text{FiO}_2 \) ratio ranging from 86 to 205 mmHg. In all patients \( \text{PaO}_2/\text{FiO}_2 \) had improved at the time of study inclusion (Table 1). Eleven patients were initially included, but one patient was excluded during the study due to low EAdi signal-to-noise ratio during rocuronium titration; as a consequence ventilation in NAVA mode was not feasible. Table 1 reports patient characteristics at study inclusion and Table 2 the relevant ventilatory settings during study protocol. Additional ventilator settings are provided in supplemental Table 1.

**Rocuronium administration**

The number of rocuronium boluses [range: 1 - 12], cumulative dose [range: 5 - 37 mg], and time [range: 6 - 60 min] required to achieve a \( V_T \leq 6 \text{ ml/kg} \) were highly variable among patients. Similarly, the dose of continuous rocuronium infusion [range: 1 - 35 mg/hr] and requirement for additional boluses [range: 0 - 10] to maintain low \( V_T \) during two hour PSV and NAVA were variable among subjects. Supplemental Table 2 and supplemental Table 3 report the details of rocuronium administration during titration and continuous infusion, respectively.

**Ventilatory response**

Baseline \( V_T \) was high in PSV-12 (9.8 ± 0.5 ml/kg) and NAVA (9.4 ± 0.6 ml/kg). Reducing PS level from 12 to 6 cmH\(_2\)O resulted in a minor reduction in \( V_T \) (PSV-6: 8.6 ± 0.6 ml/kg) and a non-significant change in respiratory rate (PSV-12: 17.7 ± 1.5 breaths/min vs. PSV-6: 20.1 ± 2.0 breaths/min; \( p = 0.16 \)). The trigger occlusion pressure in the first 100 ms (P0.1) was 3.3 ± 0.4 cmH\(_2\)O during PSV-12 and increased to 4.3 ± 0.7 cmH\(_2\)O during PSV-6 (\( p < 0.05 \)). In VCV, patients developed frequent breath-stacking with an average number of 8.0 ± 2.1 per minute (for examples see supplemental Figure 1).
Partial neuromuscular blocking in patients with high tidal volumes

Table 1. Patient characteristics at study inclusion

<table>
<thead>
<tr>
<th>#</th>
<th>Sex</th>
<th>Age (y)</th>
<th>Weight (kg)</th>
<th>Height (cm)</th>
<th>BMI (kg/m²)</th>
<th>Days ventilation</th>
<th>RASS</th>
<th>Sedation/analgesia*</th>
<th>PaO₂/FiO₂ (mmHg)</th>
<th>Admission diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>M</td>
<td>53</td>
<td>73</td>
<td>185</td>
<td>21</td>
<td>26</td>
<td>-4</td>
<td>sufentanil, midazolam</td>
<td>143</td>
<td>influenza</td>
</tr>
<tr>
<td>2</td>
<td>M</td>
<td>65</td>
<td>83</td>
<td>176</td>
<td>27</td>
<td>6</td>
<td>-4</td>
<td>-</td>
<td>145</td>
<td>pneumosepsis</td>
</tr>
<tr>
<td>3</td>
<td>M</td>
<td>61</td>
<td>86</td>
<td>186</td>
<td>25</td>
<td>4</td>
<td>-3</td>
<td>sufentanil, midazolam</td>
<td>257</td>
<td>aspiration</td>
</tr>
<tr>
<td>4</td>
<td>M</td>
<td>70</td>
<td>57</td>
<td>174</td>
<td>19</td>
<td>48</td>
<td>-4</td>
<td>sufentanil</td>
<td>187</td>
<td>interstitial lung disease</td>
</tr>
<tr>
<td>5</td>
<td>V</td>
<td>31</td>
<td>140</td>
<td>160</td>
<td>55</td>
<td>8</td>
<td>-4</td>
<td>sufentanil, propofol</td>
<td>375</td>
<td>psittacosis</td>
</tr>
<tr>
<td>6</td>
<td>M</td>
<td>63</td>
<td>76</td>
<td>176</td>
<td>25</td>
<td>4</td>
<td>-3</td>
<td>propofol</td>
<td>198</td>
<td>legionella</td>
</tr>
<tr>
<td>7</td>
<td>M</td>
<td>62</td>
<td>87</td>
<td>175</td>
<td>28</td>
<td>0.5</td>
<td>-4</td>
<td>sufentanil, midazolam</td>
<td>188</td>
<td>abdominal sepsis</td>
</tr>
<tr>
<td>8</td>
<td>M</td>
<td>61</td>
<td>73</td>
<td>176</td>
<td>24</td>
<td>24</td>
<td>-3</td>
<td>propofol</td>
<td>247</td>
<td>post-op aorta surgery</td>
</tr>
<tr>
<td>9</td>
<td>M</td>
<td>53</td>
<td>97</td>
<td>192</td>
<td>26</td>
<td>7</td>
<td>-4</td>
<td>sufentanil, propofol</td>
<td>157</td>
<td>lung contusion</td>
</tr>
<tr>
<td>10</td>
<td>M</td>
<td>72</td>
<td>64</td>
<td>167</td>
<td>23</td>
<td>3</td>
<td>-5</td>
<td>propofol</td>
<td>115</td>
<td>pneumosepsis</td>
</tr>
</tbody>
</table>

* Type of sedation/analgesia received in past 24 hours. Patient #2 received high levels of sedation up to 72 hours before inclusion but maintained low RASS due to severe kidney failure. Patient #4 received analgesia without sedation due to an encephalopathy.

Table 2. Ventilator settings

<table>
<thead>
<tr>
<th>#</th>
<th>FiO₂ (%)</th>
<th>PEEP (cmH₂O)</th>
<th>NAVA level (cmH₂O/µV)</th>
<th>PSV level (phase 3) (cmH₂O)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>50</td>
<td>10</td>
<td>0.7</td>
<td>6</td>
</tr>
<tr>
<td>2</td>
<td>50</td>
<td>10</td>
<td>0.6</td>
<td>6</td>
</tr>
<tr>
<td>3</td>
<td>35</td>
<td>6</td>
<td>1.0</td>
<td>5</td>
</tr>
<tr>
<td>4</td>
<td>45</td>
<td>6</td>
<td>0.5</td>
<td>5</td>
</tr>
<tr>
<td>5</td>
<td>30</td>
<td>12</td>
<td>1.5</td>
<td>6</td>
</tr>
<tr>
<td>6</td>
<td>45</td>
<td>12</td>
<td>1.5</td>
<td>6</td>
</tr>
<tr>
<td>7</td>
<td>45</td>
<td>10</td>
<td>1.0</td>
<td>4</td>
</tr>
<tr>
<td>8</td>
<td>35</td>
<td>12</td>
<td>0.6</td>
<td>6</td>
</tr>
<tr>
<td>9</td>
<td>40</td>
<td>14</td>
<td>0.8</td>
<td>6</td>
</tr>
<tr>
<td>10</td>
<td>75</td>
<td>14</td>
<td>1.8</td>
<td>12</td>
</tr>
</tbody>
</table>
Partial neuromuscular blocking in patients with high tidal volumes

As illustrated in Figure 2A, in phase 2, rocuronium titration resulted in 38 ± 4 % reduction in Vₜ (from 9.3 ± 0.6 to 5.6 ± 0.2 ml/kg). During titration the percentage of breaths with a Vₜ > 8 ml/kg decreased from 78 ± 11 % to 4 ± 4 %, and the percentage of breaths with a Vₜ < 6 ml/kg increased from 1 ± 1 % to 49 ± 9 % (Figure 3). Low Vₜ could be maintained in phase 3, during continuous rocuronium infusion in both NAVA and PSV (Figure 2A). There was no difference in average Vₜ between NAVA and PSV during continuous rocuronium infusion (6.6 ± 0.4 ml/kg vs. 6.5 ± 0.3 ml/kg; p = 0.91).

Reducing PSV from 12 to 6 cmH₂O in phase 1 resulted in an increase in EAdi (from 17.0 ± 2.7 to 22.8 ± 3.3 µV) and patient WOB (from 0.83 ± 0.16 to 1.21 ± 0.20 J/L) (Figure 2C-D). Pₜ, EAdi and WOB followed a similar trend compared to Vₜ in response to partial neuromuscular blockade during titration (Figure 2B-D) with, respectively, a decrease of 58 ± 5 %, 73 ± 3 % and 40 ± 9 %. During continuous rocuronium infusion (phase 3), there were no differences between NAVA and PSV for average Pₜ and EAdi, but patient WOB was slightly lower with PSV.

Rocuronium administration had no significant effect on the coefficient of variation of Vₜ (baseline: 8.4 ± 1.0 % vs. end titration: 11.1 ± 0.8 %; p = 0.06) and the coefficient of variation of EAdi (baseline: 17.1 ± 1.7 % vs. end titration: 20.0 ± 1.5 %; p = 0.33).

Figure 4 shows the effects of rocuronium titration (phase 2) on Pvent, transdiaphragmatic pressure (Pdi), esophageal pressure (Pes), respiratory rate, and minute ventilation ( VE ) and at the end of 1 hour NAVA and PSV (phase 3). Inspiratory time did not change during the titration phase (from 0.90 ± 0.14 sec in the first minute to 0.90 ± 0.22 sec in the last minute; p = 0.436), whereas expiratory time decreased from 2.58 ± 0.23 sec in the first minute to 2.14 ± 0.23 sec in the last minute.
minute \((p < 0.05)\). Dynamic lung compliance increased during phase 2 from 26 ± 2 in the first minute to 42 ± 4.7 ml/cmH\(_2\)O in the last minute \((p < 0.0001)\). Expiratory muscle activity, measured by the increase in gastric pressure during expiration, was low at start of titration \((1.3 ± 0.37\) cmH\(_2\)O\) and did not change during the study \((1.0 ± 0.47\) after titration; \(p = 0.637)\).

Figure 5 shows that short-term use (2 hrs) of partial neuromuscular blockade does not affect neuromechanical efficiency of the diaphragm, indicating the absence of the development of diaphragm fatigue.

![Figure 5](image_url)

**Figure 3.** Distribution of low and high \(V_T\) during rocuronium titration. \(V_T\) = tidal volume.

**Cardiovascular response**

Hemodynamic data from one patient was lost due to technical issues during data acquisition. Facilitating lung protective ventilation with NMBA did have some hemodynamic consequences. Mean arterial blood pressure and heart rate increased during rocuronium titration (phase 2) from 84 ± 6 to 99 ± 6 mmHg, and 83 ± 7 to 93 ± 8 beats/min, respectively (supplemental Figure 3). Mean arterial blood pressure partially returned to baseline values in phase 3. There were no correlations between RASS and delta mean arterial blood pressure \((\text{Spearman } r = -0.29,\ p = 0.437)\), and RASS and delta heart rate \((\text{Spearman } r = -0.19,\ p = 0.613)\) during rocuronium titration. There was a significant, but clinical irrelevant, decrease in \(\text{SaO}_2\) from 98 ± 0.5 to 97 ± 0.5 % during rocuronium titration \((p = 0.01)\). There were no differences in hemodynamic response between NAVA and PSV during continuous rocuronium infusion.
Partial neuromuscular blocking in patients with high tidal volumes

Figure 4. P_{vent}, P_{di}, P_{es}, respiratory rate, and $E$ during titration (phase 2) and at the end of 1 hour NAVA and PSV (phase 3). Since ventilator output and diaphragm activity are coupled with NAVA there is a decline in P_{vent} during titration. In response to the decline in $V_t$ (Figure 2A) there is an increase in respiratory rate, nevertheless $E$ drops during titration. In panel B the circles represent $P_{di}$ ($p = 0.0001$) and the squares represent $P_{es}$ ($p = 0.0013$). $p$-values relate to the titration phase. NAVA = neurally adjusted ventilatory assist; P_{di} = transdiaphragmatic pressure; P_{es} = esophageal pressure; PSV = pressure support ventilation; P_{vent} = ventilatory pressure; $E$ = minute ventilation; $V_t$ = tidal volume.
Figure 5. Neuromechanical efficiency (NME) of the diaphragm during continuous infusion of rocuronium (phase 3). There were no differences in NME over time for NAVA and PSV, and between modes.

Figure 6. PaCO₂ and pH at different time points. As a result of partial neuromuscular blockade and the consequent drop in $i_e$, there is marked increase in PaCO₂ and decrease in arterial pH. NAVA = neurally adjusted ventilatory assist; PaCO₂ = partial pressure of CO₂ in arterial blood; PSV = pressure support ventilation. * p < 0.05 compared to baseline. + p < 0.05 compared to end titration.
Blood gas values
Figure 6 shows the results of the arterial blood gas analysis at multiple time points. Briefly, there was a significant rise in PaCO$_2$ after rocuronium titration (phase 2), that further increased during the continuous rocuronium infusion (phase 3). Consequently, pH decreased during phase 2 and further decreased during phase 3. PaO$_2$ remained stable during the study (baseline: 96 ± 7 mmHg, end of NAVA (phase 3): 96 ± 6 mmHg, end of PSV (phase 3): 100 ± 8 mmHg).

Discussion
In the present proof-of-concept study we evaluated a novel strategy to deliver lung-protective ventilation while maintaining diaphragm activity in lung injury patients with high tidal volume during partial support ventilation. We found that neuromechanical uncoupling of the diaphragm by partial neuromuscular blockade 1) facilitates lung-protective ventilation as indicated by low tidal volumes and low transpulmonary pressures and 2) reduces the work of breathing, while 3) maintaining diaphragm activity. Furthermore, low V$_T$ and P$_O$ could be maintained under continuous rocuronium infusion for at least 2 hours. However, this strategy is associated with mild hemodynamic side effects, including tachycardia and elevated blood pressure, and development of hypercapnic acidosis.

Before rocuronium administration, tidal volume and transpulmonary pressures were above the limits of lung-protective ventilation under partial support ventilation. In addition to high V$_T$, most patients had relatively high EAdi (± 17 µV) and delta Pes (± -11 cmH$_2$O) compared to other studies with comparable levels of support (21, 28, 29), indicating strong spontaneous breathing efforts, and a moderate to high P0.1 (± 4 cmH$_2$O). Reducing pressure support level from 12 cmH$_2$O to 6 cmH$_2$O decreased V$_T$ only with ±1 ml/kg (still at ± 8.6 ml/kg), but increased patient work of breathing. Further reducing pressure support and eventually CPAP would result in even higher work of breathing, which may result in the development of diaphragm injury and fatigue (30-32). Appropriate setting of PSV to attain both lung-protective ventilation and physiological levels of patient work of breathing is a clinical challenge. An assist-controlled mode under these conditions resulted in severe breath stacking (supplemental Figure 1), which is associated with high V$_T$ (33). A reasonable option for the clinician would be to switch to controlled ventilation and increase the level of sedation to decrease respiratory drive. Instead, we studied the effects of partial neuromuscular blockade of the diaphragm using the acetylcholine receptor antagonist rocuronium.
Tidal volume and transpulmonary pressure

The ventilatory response to rocuronium administration was rather unpredictable among patients (supplemental Table 2). Nevertheless, we were successful in titrating $V_T \leq 6 \text{ ml/kg}$, as dictated by the protocol in all patients, supporting our hypothesis. In response to the decreased $V_T$, respiratory rate increased, but not sufficient to maintain $E$. The latter is in contrast to a study evaluating the respiratory effects of low dose NMBA in healthy subjects, where increased respiratory rate completely compensated for reduction of $V_T$ (34). Most likely in our patients sedatives did reduce sensitivity of the central respiratory centers to increased PaCO$_2$ and reduced pH (35-38). Although such pH during the continuous rocuronium administration seems to be safe from physiological perspective and may even be lung-protective (39), in patients with light sedation this may result in enhanced dyspnea sensation.

The transpulmonary pressure is the real distending pressure of the lung parenchyma. The point at which over distention of alveolar structures occurs is called the upper physiological limit of $P_t$, which has been suggested to be around 25 cmH$_2$O (40, 41). Even when plateau pressure of the ventilator is limited to 30 cmH$_2$O, high $P_t$ generated by strong spontaneous breathing efforts have been shown, in experimental studies, to worsen lung injury (4, 42, 43). In the present study, the initial high $P_t$ ($\pm 27$ cmH$_2$O), was significantly reduced to lung-protective values ($\pm 11$ cmH$_2$O) after rocuronium titration. Our measurements of $P_t$ were performed under dynamic conditions. Thus $P_t$ consists of a resistive and elastic component, where only the latter is assumed to be injurious to the lung (29). As peak flow decreased during titration of rocuronium, part of the decrease in $P_t$ can be attributed to the resistive component.

In the current study, neuromuscular blockade was titrated to achieve a $V_T$ of 6 ml/kg, although the optimal lung-protective $V_T$ and $P_t$ under assisted ventilation is subject of debate. In mild to moderate ARDS beneficial effects of spontaneous breathing efforts have been demonstrated in clinical studies (15-17). In particular, redistribution of ventilation due to alveolar recruitment of dependent lung areas may reduce lung injury (15). However, patient breathing efforts may promote lung injury in severe ARDS through different mechanisms (43, 44). First, large negative swings in $P_t$ are more likely to occur in severe ARDS (43). Second, development of pendelluft may result in inhomogeneous stretch of dependent lung areas and may additionally contribute to lung injury (44). Accordingly, whether spontaneous breathing efforts in patients with injured lungs are beneficial or damaging depends on several factors, as outlined above. This concept has recently been elegantly reviewed by Yoshida and colleagues (23). Although the optimal $V_T$ and $P_t$ are debated in patients with ARDS under partial support ventilation, titration of a neuromuscular blocker facilitates precise control of these variables.
Partial neuromuscular blocking in patients with high tidal volumes

Diaphragm activity
High dose non-depolarizing NMBAs (such as rocuronium) competitively inhibit the postsynaptic acetylcholine receptors of the neuromuscular motor endplate, preventing depolarization and as such force generation (45). Using low dose rocuronium as in the present study allows more subtle modulation of $E_{Adi}$ and $P_{di}$. In other words, strong spontaneous breathing efforts were abolished, but a significant degree of diaphragm activity was maintained. In animal studies, maintaining spontaneous respiratory efforts during mechanical ventilation alleviated the development of VIDD (18-20). As to whether the levels of diaphragm activity we obtained are sufficient to prevent or alleviate the development of VIDD is unknown, but after titration of rocuronium, near physiological levels of diaphragm activity were attained (Figure 2).

Administration of low dose muscle rocuronium induces acute muscle weakness. Therefore, a possible concern of this strategy is the development of respiratory muscle fatigue. Moreover, non-depolarizing neuromuscular blockers, such as rocuronium, tend to produce less blockade at fast-twitch (fatigue resistant) than slow-twitch fibers (46, 47). Throughout the 2 hours of continuous infusion the stability of $V_p$, $E_{Adi}$ and $P_{di}$ confirms it is feasible to maintain partial paralysis. $E_{Adi}$ represents the spatio-temporal summation of motor unit action potentials, which in turn translates into muscle-tension and $P_{di}$. Thus, the stable NME (Figure 5) contradicts that the muscle tension developed for a given $E_{Adi}$ was reduced, virtually excluding the development of contractile fatigue of the diaphragm. Whether prolonged administration of low dose rocuronium results in fatigue or injury of the diaphragm remains to be investigated.

It should be emphasized that partial neuromuscular blockade induces neuromechanical uncoupling of the diaphragm and therefore the reduction in $E_{Adi}$ does not reflect a decreased neural respiratory drive, which is most likely increased. Studying respiratory drive under neuromuscular blockade would require measuring activity of the central respiratory centers or the phrenic nerve which is not feasible in ICU patients.

Cardiovascular response
Titration of rocuronium resulted in a mild increase in arterial blood pressure and heart rate. Supposedly, the cardiovascular response to partial neuromuscular blockade could be mediated through several pathways. First, direct cardiovascular effects of rocuronium administration (2 to 4 times the ED95) have been studied previously (48-50). The general conclusion from these studies in anesthetized and ventilated patients is that rocuronium has virtually no direct effects on hemodynamic variables. Moreover, we used much lower doses of rocuronium. Second, patients developed hypercapnic
acidosis in phase 2 and 3 of the study. The integrated hemodynamic response to hypercapnic acidosis during complete paralysis, as part of a lung-protective approach in ARDS, has been well described, and may be characterized by an increase in cardiac output, heart rate, and stroke volume, a decrease in systemic vascular resistance, and no effect on mean arterial blood pressure (51-53). This sympathetically activated response by hypercapnic acidosis is supported by observations of higher perfusion to the adrenal glands and production of catecholamines in the state of hypercapnic acidosis (54, 55). Previously, it was demonstrated that rapid induction of hypercapnic acidosis by reducing V̇ in patients with ARDS is associated with impaired right ventricular function and reduced cardiac index (56). In contrast to our study, in that study MAP decreased. Third, physical stress in response to increased dyspnea sensation may increase heart rate and blood pressure. However, patients in our study were moderately to deeply sedated, no physical signs of stress (anxiety, agitation, diaphoresis) were observed, and we found no correlation between sedation depth and the changes in heart rate and blood pressure during rocuronium titration. In addition, both heart rate and systolic blood pressure decreased again during phase 3 (continuous infusion) to baseline levels. Fourth, increased central neural drive to the diaphragm during partial neuromuscular blockade may activate the cardiovascular response in parallel. This feed-forward neural mechanism in response to physical activity is termed ‘central command’ (57). Experimental studies to assess the influence of central command on the cardiovascular response have demonstrated that partial neuromuscular blockade during static and dynamic exercise leads to a concomitant increase in heart rate and mean arterial pressure (58-61). This was suggested to be directed by way of increased plasma catecholamine levels (60). To minimize hemodynamic consequences in the future, the increase in PaCO₂ could be made less rapid by titrating rocuronium more slowly.

Clinical implications
Our results may have potential future clinical implication with respect to patients recovering from ARDS with high tidal volume and high transpulmonary pressure during partial support ventilation. Recently, we have shown that partial support modes allow lung-protective ventilation in sedated patients with stable ARDS (PaO₂/FiO₂ 144 ± 12 mmHg) (21). However, in patients with strong spontaneous breathing efforts this may result in high V̇ and lung distending pressures. In the present study, we demonstrated the feasibility of partial neuromuscular blockade to facilitate lung-protective ventilation during partial support ventilation while preserving diaphragm activity. Because the current study is only a proof of concept, it is too early for clinical application. The long-term effects of partial neuromuscular blockade on respiratory muscle function need to be studied. In addition, titration with rocuronium is complicated and requires
continuous observation of the patients’ respiratory muscle activity to adjust rocuronium dose. Several tools are available to monitor respiratory muscle function (62). All patients in the present study were sedated. One of the intended effects of partial neuromuscular blockade is to reduce the need for sedation. Future studies should carefully evaluate whether partial neuromuscular blockade is tolerated by patients under light or no sedation. As mentioned before, progressive partial paralysis has been studied in healthy volunteers without sedation (26, 34, 35, 63-69).

Study limitations
This study has limitations. First, our study is a single-center, proof-of-concept study, conducted in a selected and small group of patients. This limits the interpretation of our data. Moreover, the relative short study period of 2 hours of partial neuromuscular blockade might not be enough to reflect the consequences for patients who are ventilated for longer times.

Second, only physiological markers associated with the development of lung injury (V_t and P_L) and diaphragm function (EAdi, Pdi) were evaluated. Future studies should also assess the effect of this strategy on direct markers for lung injury and diaphragm injury (i.e. histology, inflammation).

Third, prior to titration of neuromuscular blockade we did not evaluate the effect of different PEEP levels on breathing effort. Increasing PEEP level will increase end-expiratory lung volume (EELV) (70), which may result in a reduction in neural respiratory drive via the positive effects of lung recruitment (71). Nevertheless, modulation in respiratory drive by increase in EELV is modest compared to the reductions in diaphragmatic force that were required to achieve 6 ml/kg in our study.

Fourth, only the diaphragm was evaluated in this study, so we are unaware of the effects of partial neuromuscular blockade on other respiratory and peripheral muscles. Previous studies demonstrated that the diaphragm is less susceptible to neuromuscular blockade compared to peripheral muscle, called the respiratory sparing effect (34, 67, 68). Therefore, it is likely that peripheral muscles were at least equally affected as the diaphragm.

In conclusion, the present proof-of-concept study demonstrates that partial neuromuscular blockade facilitates lung-protective ventilation during partial ventilatory support, while maintaining diaphragm activity. These findings are of potential interest for patients recovering from ARDS with spontaneous breathing efforts under modest levels of partial ventilatory assist leading to high tidal volumes. Further research should focus on better control of hemodynamic effects and understanding of the long-term consequences on muscular function and dyspnea.
References


Partial neuromuscular blocking in patients with high tidal volumes


Supplemental material

Supplemental methods

Sedation protocol

Patients were sedated during the study, although not all patients received sedatives at the time of the study execution (see Table 1 of the main manuscript). Patient #2 received high levels of sedation up to 72 hours before inclusion but was still sedated, probably due to severe kidney failure. Patient #4 received analgesics (sufentanil) without sedatives due to an encephalopathy. A senior critical care physician and nurse not involved in the study were present at the bedside to assure comfort of the patient. A bolus of either propofol or midazolam could be administered when deemed appropriate by the nurse, according to our clinical protocol for ICU sedation. A bolus was administered in 4 of the patients included in this study during rocuronium titration.

Titration protocol

The goal of rocuronium titration was to partially block diaphragmatic force to obtain a $V_T$ of 6 ml/kg. Therefore, rocuronium was titrated on $V_T$ and not guided by the train-of-four technique. Following every bolus of rocuronium (2 to 5 mg) an average tidal volume of 10 breaths was calculated to evaluate whether the target of 6 ml/kg was reached. If the target was not reached a subsequent bolus was given. This was repeated until a $V_T$ of 6 ml/kg was reached.

Data acquisition

Electrical activity of the diaphragm (EAdi), esophageal pressure (Pes) and gastric pressure (Pga) were measured with a multi-electrode esophageal catheter with double balloons (Neurovent Research Inc., Toronto, Canada). EAdi refers to a method using a standard electrode, acquisition, and analysis system to overcome signal filtering and processing effects when quantifying the electromyogram of the diaphragm (72). Specifically, EAdi in the present study refers to the calibration in µV made for the commercially available Servo-i ventilator (Maquet Critical Care, Sölna, Sweden). Details on catheter positioning were published previously by our group (21, 73). Flow, ventilatory pressure (Pvent), Pes, Pga, and EAdi tracings were acquired synchronously and continuously during all three phases of the study protocol as described previously (21). Transdiaphragmatic (Pdi) and transpulmonary pressure (P′) were calculated as the difference between Pga and Pes, and Pvent and Pes, respectively.
Partial neuromuscular blocking in patients with high tidal volumes

An arterial catheter was used for continuous blood pressure monitoring and blood sampling. Heart rate and rhythm were monitored using a standard 5-lead electrocardiography system. Pulse oximetry was used to monitor arterial oxygen saturation (SaO\textsubscript{2}). Blood pressure, heart rate and SaO\textsubscript{2} were sampled every 30 seconds from a patient monitor (MP50, Philips, Eindhoven, The Netherlands) by an in-house developed data acquisition system.

Data analysis
All tracings were analyzed off-line using a software routine developed for Matlab (R2014b, Mathworks, Natick, MA, USA). Variables from the respiratory tracings were analyzed on a breath-by-breath basis. For phase 1 a 2-min period at the end of each mode was selected for analysis. The duration of phase 2 is different per patient, therefore data were analyzed at seven points in time: the first and last minute and five 1-min periods taken at equal time intervals in between. For phase 3 a 2-min period was selected for analysis every ten minutes of each mode during 1 hour. For each breath, we defined the following variables during the inspiratory phase:

- \( V_T \): as the integral of the inspiratory flow over time.
- \( \Delta P_{vent} \): as maximal change from PEEP level.
- \( \Delta \) and absolute \( P_L \). For \( \Delta P_L \) baseline was defined as transpulmonary pressure at start of inspiration.
- \( \Delta P_{di} \): as change from baseline. Baseline was defined as Pdi at start of inspiration.
- \( P_{0.1} \) was calculated from Pes according to a previously described method (74)
- Peak EAdi
- Neuromechanical efficiency (NME), as the ratio between Pdi and EAdi.
- Work-of-breathing (WOB), as calculated from the Campbell diagram (75).
- Dynamic compliance (\( C_{dyn} \)), as \( V_T / \Delta P_L \).

Breath-by-breath variability was assessed by calculating the coefficient of variation (CV; [standard deviation/mean]*100) for \( V_T \) and EAdi. Blood pressure, heart rate and SaO\textsubscript{2} were averaged over the same time intervals as previously mentioned.
Supplemental references


Partial neuromuscular blocking in patients with high tidal volumes

Supplemental Table 1. Additional ventilator settings

<table>
<thead>
<tr>
<th>#</th>
<th>Assist-volume control</th>
<th>Pressure support</th>
<th>NAVA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>I:E ratio</td>
<td>Pause time (% insp. time)</td>
<td>Insp. trigger (l/min)</td>
</tr>
<tr>
<td>1</td>
<td>1:2</td>
<td>0</td>
<td>1.4</td>
</tr>
<tr>
<td>2</td>
<td>1:1.5</td>
<td>10</td>
<td>1.4</td>
</tr>
<tr>
<td>3</td>
<td>1:2</td>
<td>10</td>
<td>1.4</td>
</tr>
<tr>
<td>4</td>
<td>1:2</td>
<td>0</td>
<td>1.0</td>
</tr>
<tr>
<td>5</td>
<td>1:2</td>
<td>10</td>
<td>1.4</td>
</tr>
<tr>
<td>6</td>
<td>1:2</td>
<td>0</td>
<td>0.6</td>
</tr>
<tr>
<td>7</td>
<td>1:2</td>
<td>10</td>
<td>1.6</td>
</tr>
<tr>
<td>8</td>
<td>1:2</td>
<td>0</td>
<td>1.4</td>
</tr>
<tr>
<td>9</td>
<td>1:2</td>
<td>0</td>
<td>1.6</td>
</tr>
<tr>
<td>10</td>
<td>1:2</td>
<td>10</td>
<td>1.6</td>
</tr>
</tbody>
</table>

All patients were ventilated with a Servo-i ventilator (Maquet Critical Care, Sölna, Sweden) with a MR850 humidification device (Fisher & Paykel Healthcare Limited, Auckland, New Zealand). As a secondary source NAVA also employs the flow trigger, set as shown in the table, which operates in combination with the neural trigger on a first-come-first-served basis. Expiratory triggering in NAVA starts when the EAdi decreases below 70% for normal and high EAdi signals (40% for low EAdi signals) of the peak value.

Supplemental Table 2. Rocuronium administration (titration phase)

<table>
<thead>
<tr>
<th>#</th>
<th>Bolus (#)</th>
<th>Total bolus (mg)</th>
<th>Bolus (mg/#)</th>
<th>Time (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>11</td>
<td>34.0</td>
<td>3.1</td>
<td>33</td>
</tr>
<tr>
<td>2</td>
<td>1</td>
<td>5.0</td>
<td>5.0</td>
<td>6</td>
</tr>
<tr>
<td>3</td>
<td>3</td>
<td>15.0</td>
<td>5.0</td>
<td>12</td>
</tr>
<tr>
<td>4</td>
<td>2</td>
<td>7.5</td>
<td>3.8</td>
<td>10</td>
</tr>
<tr>
<td>5</td>
<td>6</td>
<td>31.0</td>
<td>5.2</td>
<td>42</td>
</tr>
<tr>
<td>6</td>
<td>6</td>
<td>22.0</td>
<td>3.7</td>
<td>36</td>
</tr>
<tr>
<td>7</td>
<td>5</td>
<td>14.0</td>
<td>2.8</td>
<td>14</td>
</tr>
<tr>
<td>8</td>
<td>5</td>
<td>30.0</td>
<td>6.0</td>
<td>23</td>
</tr>
<tr>
<td>9</td>
<td>12</td>
<td>37.0</td>
<td>3.1</td>
<td>60</td>
</tr>
<tr>
<td>10</td>
<td>4</td>
<td>14.0</td>
<td>3.5</td>
<td>12</td>
</tr>
<tr>
<td>mean</td>
<td>6</td>
<td>21.0</td>
<td>4.1</td>
<td>25</td>
</tr>
<tr>
<td>SEM</td>
<td>1</td>
<td>3.6</td>
<td>0.3</td>
<td>6</td>
</tr>
</tbody>
</table>
**Supplemental Table 3.** Continuous rocuronium infusion and additional boluses (phase 3)

<table>
<thead>
<tr>
<th>#</th>
<th>Continuous <em>(mg/h)</em></th>
<th>Bolus (#)</th>
<th>Total bolus (mg)</th>
<th>Bolus (mg/#)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>35</td>
<td>9</td>
<td>27</td>
<td>3.0</td>
</tr>
<tr>
<td>2</td>
<td>14</td>
<td>0</td>
<td>0</td>
<td>0.0</td>
</tr>
<tr>
<td>3</td>
<td>5</td>
<td>3</td>
<td>10</td>
<td>3.3</td>
</tr>
<tr>
<td>4</td>
<td>8</td>
<td>3</td>
<td>6</td>
<td>2.0</td>
</tr>
<tr>
<td>5</td>
<td>26</td>
<td>10</td>
<td>24</td>
<td>2.4</td>
</tr>
<tr>
<td>6</td>
<td>10</td>
<td>6</td>
<td>19</td>
<td>3.2</td>
</tr>
<tr>
<td>7</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0.0</td>
</tr>
<tr>
<td>8</td>
<td>18</td>
<td>10</td>
<td>39</td>
<td>3.9</td>
</tr>
<tr>
<td>9</td>
<td>33</td>
<td>4</td>
<td>12</td>
<td>3.0</td>
</tr>
<tr>
<td>10</td>
<td>11</td>
<td>2</td>
<td>4</td>
<td>2.0</td>
</tr>
<tr>
<td>mean</td>
<td>16</td>
<td>5</td>
<td>14</td>
<td>2.3</td>
</tr>
<tr>
<td>SEM</td>
<td>4</td>
<td>1</td>
<td>4</td>
<td>0.4</td>
</tr>
</tbody>
</table>

* average value of 2 hours
Supplemental Figure 1.
Breath-stacking caused by reversed triggering in patients 2, 5, 7 and 9 during volume control ventilation. Red circles represent early termination of expiratory flow causing breath-stacking. EAdi = electrical activity of the diaphragm; Pvent = ventilatory pressure.
Supplemental Figure 2. Progress of absolute $P_L$ and WOB (J/min) during the different phases of the study protocol. Without neuromuscular blockade absolute $P_L$ was high in all three assisted modes. Rocuronium titration resulted in a gradual, but strong, decline of both parameters. During continuous rocuronium infusion more or less stable values could be obtained in both NAVA and PSV. $p < 0.05$ for NAVA vs. PSV-6 and PSV-12. NAVA = neurally adjusted ventilatory assist; $P_L$ = transpulmonary pressure. PSV = pressure support ventilation; WOB = work of breathing.
Supplemental Figure 3. Cardiovascular response during rocuronium titration (phase 2) and at the end of 1 hour NAVA and PSV (phase 3). ABP = arterial blood pressure; HR = heart rate; NAVA = neurally adjusted ventilatory assist; PSV = pressure support ventilation.
Chapter 9

The calcium sensitizer levosimendan improves human diaphragm function

Jonne Doorduin, Christer A. Sinderby, Jennifer Beck, Dick F. Stegeman, Hieronymus W.H. van Hees, Johannes G. van der Hoeven, Leo M.A. Heunks

Abstract

Rationale: Acquired diaphragm muscle weakness is a key feature in several chronic diseases including chronic obstructive pulmonary disease, congestive heart failure, and difficult weaning from mechanical ventilation. However, no drug is available to improve respiratory muscle function in these patients. Recently, we have shown that the calcium sensitizer levosimendan enhances force generating capacity of isolated diaphragm fibers.

Objectives: To investigate the effects of the calcium sensitizer levosimendan on in vivo human diaphragm function.

Methods: In a double-blind randomized crossover design, 30 healthy subjects performed two identical inspiratory loading tasks. After the first loading task, subjects received levosimendan (40 µg/kg bolus followed by 0.1/0.2 µg/kg/min continuous infusion) or placebo. Transdiaphragmatic pressure, diaphragm electrical activity and their relationship (neuro-mechanical efficiency) were measured during loading. Magnetic phrenic nerve stimulation was performed before the first loading task and after bolus administration to assess twitch contractility. Center frequency of diaphragm electrical activity was evaluated to study the effects of levosimendan on muscle fiber conduction velocity.

Measurements and Main results: The placebo group showed a 9% (P=0.01) loss of twitch contractility after loaded breathing, whereas no loss in contractility was observed in the levosimendan group. Neuro-mechanical efficiency of the diaphragm during loading improved by 21% (P<0.05) in the levosimendan group. Baseline center frequency of diaphragm electrical activity was reduced after levosimendan administration (P<0.05).

Conclusions: The calcium sensitizer levosimendan improves both neuro-mechanical efficiency and contractile function of the human diaphragm. Our findings suggest a new therapeutic approach to improve respiratory muscle function in patients with respiratory failure.
Introduction

Impaired force generation of the respiratory muscles has been recognized in a variety of diseases including chronic obstructive disease (COPD), congestive heart failure and in the critically ill (1-7). The pathophysiological substrate of diaphragm dysfunction in these disorders is multifactorial and includes muscle fiber atrophy and contractile protein dysfunction (3, 8-11). We found that diaphragm muscle fibers of patients with COPD display reduced sensitivity of the contractile proteins to calcium (9, 12). In other words more calcium is needed to develop the same amount of force as in the non-COPD diaphragm, resulting in impaired contractile efficiency of the diaphragm muscle. Subsequent studies have shown that calcium sensitivity is also reduced in the diaphragm of animal models for congestive heart failure and prolonged mechanical ventilation (2, 13). Despite a better understanding of respiratory muscle dysfunction in chronic diseases, currently no drug is available to improve respiratory muscle function in humans.

Levosimendan is a clinically used calcium sensitizer, approved to enhance cardiac contractility in patients with acute heart failure. Clinical studies have shown that levosimendan improves cardiac function in these patients (14). Levosimendan enhances the binding of calcium to troponin C, thereby improving the responsiveness of myofilaments to calcium. Accordingly, a greater amount of force is generated for the same level of cytosolic calcium, resulting in enhanced contractile efficiency. Besides calcium sensitizing, levosimendan has vasodilatory properties mediated by the opening of the ATP-sensitive potassium (K\textsubscript{ATP}) channels (15).

Recently, we have shown that levosimendan also enhances calcium sensitivity of permeabilized muscle fibers obtained from the human diaphragm, including patients with COPD (12). However, the effects of calcium sensitizing on the human diaphragm \textit{in vivo} have not been studied. Based on our \textit{in vitro} data (12) we hypothesize that levosimendan improves contractile function of the human diaphragm \textit{in vivo} through calcium sensitizing. In addition, levosimendan would decrease muscle fiber conduction velocity of the diaphragm, through its effects on K\textsubscript{ATP} channels in diaphragm fibers. We tested this hypothesis in a double-blind placebo controlled crossover design in healthy subjects performing inspiratory loading tasks. Some of the results of this study have been previously reported in the form of an abstract (16).
Methods

We enrolled 30 healthy volunteers in this trial. The protocol was approved by the ethical committee of the Radboud University Nijmegen Medical Centre and registered at ClinicalTrials.gov (NCT01101620). All subjects gave their informed consent.

Esophageal catheter
Diaphragm EMG (EMGdi), esophageal (Pes) and gastric (Pga) pressure were obtained with a multi-electrode esophageal catheter with two balloons; see the supplemental material for details. Pdi was calculated as Pga-Pes.

Magnetic stimulation and maximal inspiratory effort
Cervical magnetic stimulation of the phrenic nerves was performed to measure twitch Pdi (Pdi$_{tw}$) and compound muscle action potential of the diaphragm (CMAPdi). See the supplemental material for stimulation protocol details.

Maximal voluntary Pdi (Pdi$_{max}$) was measured as mean Pdi in the first second during a maximal inspiratory effort against a closed valve at functional residual capacity.

Inspiratory loading task
Each subject performed two identical loading tasks, before and after administration of study medication. Sitting in upright position, with uncast abdomen subjects breathed through a mouthpiece wearing a nose clip. Subjects performed intermittent inspiratory maneuvers of 10 second against a closed valve (near-isometric contractions) followed by 7 seconds of unloaded breathing. Duty cycle was imposed by a sound signal and subjects were asked to target 40% of Pdi$_{max}$. Visual feedback of Pdi was provided. Total loading task duration was 10 minutes. During loading, EMGdi and Pdi were recorded continuously, as well as in unloaded conditions approximately 5 minutes before and after loading. Respiratory effort sensation was scored with a Borg scale (range 6-20) at one, three, six, and nine minutes into loading.

Experimental protocol
The protocol is presented in Figure 1. Pdi$_{tw}$ and Pdi$_{max}$ were measured and followed by the first loading task and 30 minutes of unloaded breathing. After randomization subjects received study medication, either levosimendan bolus (40 µg/kg bodyweight, iv) or an equal volume of placebo in 10 minutes. Pdi$_{tw}$ and Pdi$_{max}$ measurements were repeated and followed by 30 minutes of continuous levosimendan (0.1 µg/kg bodyweight/min, iv) or placebo infusion. A second loading task was performed while infusing levosimendan (0.2 µg/kg/min, iv) or placebo.
Heart rate, end tidal carbon dioxide (etCO₂) and peripheral oxygen saturation (SpO₂) were monitored continuously. Blood pressure was measured non-invasively every 10 minutes and during bolus administration each minute. In six subjects cardiac output was determined by transthoracic echocardiography directly before and after bolus administration.

**Figure 1.** Schematic description of the protocol. Abbreviations: Pdi_{tw} = twitch transdiaphragmatic pressure; Pdi_{max} = maximum transdiaphragmatic pressure.
Data Analysis and Statistics
Measurement variables were analyzed offline in Matlab R2009b (The Mathworks, Natick, MA); see the supplemental material for details. The ratio of mean inspiratory Pdi and EMGdi amplitude was calculated as a measure of the neuro-mechanical efficiency of the diaphragm. Changes in muscle fiber conduction velocity were evaluated using the power spectrum center frequency (CFdi) of the EMGdi; see the supplemental material for details. Comparisons were made with the appropriate t-test. Changes over time during loaded breathing were analyzed using repeated measures ANOVA. Values are means ± SEM, and \( P < 0.05 \) was considered significant. Statistical analyses were performed with SPSS 16.0 (SPSS, Chicago, IL, USA).

Results
Baseline characteristics of the subjects (male/female 23/7; age 22 ± 0.4 yrs; bodyweight 74 ± 1 kg; and body mass index 23 ± 0.4 kg/m\(^2\)) were not different between the placebo and levosimendan group. All subjects (n=30) received study medication as dictated by the protocol. One subject was excluded due to technical problems with the esophageal catheter. Two subjects were not able to maintain targeted Pdi (less than 30% of Pdimax) during the first loading task and were excluded from data analysis. The remaining subjects (placebo n=13 and levosimendan n=14) were able to keep their targeted Pdi during both loading tasks (44 ± 1% and 43 ± 2%), indicating compliance to the protocol. SpO\(_2\) was above 96% in all subjects during the study, without supplemental oxygen. There were no differences in etCO\(_2\) between the placebo and levosimendan group during loaded breathing. Furthermore, there were no serious adverse events during the experimental protocol. One subject, receiving levosimendan, experienced a mild degree of nausea after completion of the study.

Diaphragm contractility
Cervical magnetic stimulation of the phrenic nerve resulted in reproducible twitch pressures in all subjects. Data from magnetic stimulation and maximal inspiratory maneuvers, as well as hemodynamic data, are given in Table 1. There was no significant difference in Pdi\(_{tw}\) and Pdi\(_{max}\) between the placebo and levosimendan group before study medication.

Figure 2 shows a decreased contractile response of the diaphragm after the first loading task in a subject receiving placebo. On average, loaded breathing in subjects receiving placebo resulted in significant reductions in Pdi\(_{tw}\) (-9 ± 3 %, \( P=0.01 \) and
Levosimendan improves human diaphragm function

$\text{Pdi}_{\text{max}} (-5\% \pm 3, P<0.05)$. The group receiving levosimendan after the first period of loaded breathing revealed no significant decrease in $\text{Pdi}_{\text{tw}}$ and $\text{Pdi}_{\text{max}}$ (Figure 3 and Table 1).

**Table 1.** Effect of loading and levosimendan on the diaphragm and hemodynamics

<table>
<thead>
<tr>
<th></th>
<th>Placebo (n=13)</th>
<th>Levosimendan (n=14)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>baseline</td>
<td>after loading + bolus administration</td>
</tr>
<tr>
<td>$\text{Pdi}_{\text{tw}}$ (cmH$_2$O)</td>
<td>35 ± 2</td>
<td>32 ± 2 *</td>
</tr>
<tr>
<td>$\text{CMAPdi}$ (mV)</td>
<td>1.3 ± 0.2</td>
<td>1.3 ± 0.2</td>
</tr>
<tr>
<td>$\text{Pdi}_{\text{max}}$ (cmH$_2$O)</td>
<td>130 ± 8</td>
<td>122 ± 7 *</td>
</tr>
<tr>
<td>$\text{EMGdi}_{\text{max}}$ (µV)</td>
<td>76 ± 9</td>
<td>81 ± 10</td>
</tr>
<tr>
<td>MAP (mmHg)</td>
<td>92 ± 2</td>
<td>89 ± 4</td>
</tr>
<tr>
<td>HR (bpm)</td>
<td>71 ± 3</td>
<td>73 ± 3</td>
</tr>
<tr>
<td>CO (l/min)</td>
<td>4.5 ± 1.3</td>
<td>4.6 ± 0.4</td>
</tr>
</tbody>
</table>

Diaphragm contractility and hemodynamic variables at baseline and after loading + bolus administration. Twitch transdiaphragmatic pressure ($\text{Pdi}_{\text{tw}}$), compound muscle action potential of the diaphragm ($\text{CMAPdi}$), maximum transdiaphragmatic pressure ($\text{Pdi}_{\text{max}}$), and maximum electromyographic activity ($\text{EMGdi}_{\text{max}}$) were measured before the first loading task and after bolus administration of study medication. Mean arterial pressure (MAP), heart rate (HR) and cardiac output (CO) were measured directly before and after bolus administration. Cardiac output has only been measured in n=2 (placebo group) and n=4 (levosimendan group). Data are presented as mean ± SEM. Abbreviations: * Significantly different from before study medication (P < 0.05).

**Neuro-mechanical efficiency of the diaphragm**

In Figure 4, representative tracings of $\text{EMGdi}$ and $\text{Pdi}$ are presented for the first loading task (before study medication) and the second loaded task (with study medication) for both groups. Neuro-mechanical efficiency of the diaphragm ($\text{Pdi}/\text{EMGdi}$) in the first loading task was not different between the placebo and levosimendan group (both 1.3 ± 0.2 cmH$_2$O/µV) and remained stable throughout the loading task (Figure 5). Neuro-mechanical efficiency during unloaded breathing and during the second loading task was improved by 21% (P<0.05, compared to the first loading task) in the levosimendan group, whereas neuro-mechanical efficiency was not affected in the placebo group (Figure 5). The improved neuro-mechanical efficiency in subjects receiving levosimendan sustained throughout the entire loading task.
Figure 2. (A) the compound muscle action potential of the diaphragm (CMAPdi) and (B) twitch transdiaphragmatic pressure (Pdiₜw) elicited by cervical magnetic stimulation before (black solid line) and after the first loading task (blue striped line) in a subject receiving placebo. In response to equal diaphragm activation (i.e. equal CMAPdi), there is decreased contractility after loaded breathing. Note the difference in scale of the x-axis between figure A and B.

Figure 3. Percentage change in transdiaphragmatic twitch pressure (Pdiₜw) from baseline after loading + bolus administration in the placebo (filled squares) and levosimendan (open circles) group. Data are presented as mean ± SEM (placebo n=13, levosimendan n=14). *Significantly different from baseline (P=0.01) and levosimendan group after loading + bolus administration (P=0.01).
Levosimendan improves human diaphragm function

Figure 4. Diaphragm electromyography (EMGdi) and transdiaphragmatic pressure (Pdi) during the first (black) and second loading task (blue) in a subject receiving placebo (A and B) and levosimendan (C and D). Subjects were asked to target 40% of Pdimax. In the subject receiving placebo there is no change in EMGdi (i.e. neural activation), whereas the subject receiving levosimendan shows decreased neural activation and thus increased neuromechanical efficiency of the diaphragm.

Center frequency of diaphragm electrical activity
During both loaded breathing protocols, CFdi decreased over time ($P<0.001$; Figure supplemental Figure 1) in both groups. Administration of levosimendan resulted in a downward shift in baseline CFdi during the second loaded breathing protocol ($P<0.05$; supplemental Figure 1), whereas placebo did not affect CFdi.

Respiratory effort sensation
Respiratory effort sensation increased over time ($P<0.001$) during loaded breathing, until a score of 16. Neither levosimendan nor placebo affected respiratory effort sensation.
Discussion

The present study is the first to evaluate the effect of the calcium sensitizer levosimendan on human diaphragm function in vivo. In this double-blinded and randomized study an intervention of loaded breathing resulted in significant loss of diaphragm contractility in the placebo group but not in the levosimendan group, suggesting that levosimendan restored diaphragm contractility. Loaded breathing following drug administration was associated with improved neuro-mechanical efficiency of the diaphragm in the levosimendan group. Furthermore, levosimendan reduced baseline center frequency of diaphragm electrical activity. Levosimendan was well tolerated with negligible side effects.

Calcium sensitivity and muscle fiber conduction velocity: effects of levosimendan

Skeletal muscle force develops as intracellular calcium rises and binds to troponin C, resulting in conformational changes in the troponin complex, allowing interaction
between actin and myosin to form force generating cross-bridges. Reuptake of calcium from the cytoplasm into the sarcoplasmic reticulum is a high energy consuming process (up to 40% of total energy expenditure). Thus, reduced sensitivity of the troponin complex for calcium requires higher levels of cytoplasmic calcium to generate the same amount of force, requiring higher energy consumption and elevated CO₂ production.

Levosimendan belongs to a relatively new class of drugs, the calcium sensitizers and is approved in more than 40 countries worldwide to improve cardiac function in patients with acute heart failure (14). Although the loading dose of levosimendan in this study is higher than recommended for the treatment of acute heart failure (14), the total dose of levosimendan administered is lower than used in clinical practice.

Calcium sensitization by levosimendan occurs through stabilization of the interaction between calcium and the troponin complex during muscle activation (17). Recently, we have shown that levosimendan improves in vitro calcium sensitivity of human diaphragm (skeletal) muscle fibers as well (12, 18). Improvement in calcium sensitivity will affect contractile efficiency of the diaphragm (19). In the current study, loaded breathing reduced Pdiₜₜw in placebo treated subjects (Table 1 and Figure 3), which is consistent with previous data by Laghi and colleagues (20). However, levosimendan reversed the development of fatigue, as Pdiₜₜw after loading in these subjects was not different from baseline. Neuro-mechanical efficiency of the diaphragm during loaded breathing, expressed as Pdi/EMGdi ratio, was enhanced by 21% following administration of levosimendan (Figure 5). A 21% improvement in the ability to generate inspiratory pressure for a given neural respiratory effort should be considered a clinically relevant improvement in contractility. In vitro studies have shown that impaired calcium sensitivity of force generation develops during muscle loading and may contribute to contractile failure (21). This is in line with the data from the current study which show that levosimendan reverses the effects of diaphragm force loss (Table 1 and Figure 3).

Administration of levosimendan resulted in a downward shift of the CFdi curve during loaded breathing (supplemental Figure 1). The decrease in CFdi over time indicates the development of reduced muscle fiber conduction velocity; see the supplemental material for details. Reduced muscle fiber conduction velocity is an increased difficulty of muscle fiber action potentials to propagate along the sarcolemma and into T tubuli (22). Therefore, the downward shift in baseline CFdi in subjects receiving levosimendan most likely reflects a baseline decrease in muscle fiber conduction velocity. These data are in line with the established effects of levosimendan on K⁺ ATP channel activation (15); and that extracellular [K⁺] accumulation contributes to reduced muscle fiber conduction velocity (23, 24). Since administration
of levosimendan improves neuro-mechanical efficiency of the diaphragm with 21%, it is unlikely that extracellular [K⁺] accumulation results in functional impairment of the diaphragm. Moreover, preserving a hyperpolarized sarcolemma by activating KₚATP channels might even play a myoprotective role under conditions of metabolic stress (25).

All in all, enhancing calcium sensitivity with levosimendan appears a rational and effective approach to improve respiratory muscle function, in particular in patients with imminent respiratory failure.

**Clinical implications**

Reduced force generation of the respiratory muscles has been demonstrated in numerous disorders, including chronic obstructive disease (COPD), congestive heart failure, pulmonary hypertension and patients on mechanical ventilation (1-7). Today, there are no pharmacological interventions available to improve respiratory muscle function.

Aubier and colleagues have previously shown that dopamine (26) and aminophylline (27) improve contractility of the human diaphragm, although the effects of aminophylline are controversial (28). Neither dopamine, nor aminophylline have been used in clinical practice to optimize respiratory muscle function, probably due to the small therapeutic window, with risk of severe side effects (aminophylline, dopamine) and the very short half life time (dopamine). Instead, the active levosimendan metabolite OR-1896 has a half life time of 70-80 hours resulting in clinical effect up to one week after 24 hour infusion (29). More importantly, both aminophylline and dopamine improve contractility by elevating intracellular calcium concentration, resulting in an increased ATP utilization. As discussed previously (12), inorganic phosphate accumulation contributes to the development of muscle fatigue and depresses calcium sensitivity (30). In addition, elevated energy expenditure increases CO₂ production, requiring a higher level of ventilation. This limits the clinical utility of these drugs for the improvement of respiratory muscle function in patients with imminent ventilatory failure. Rather, calcium sensitizers improve muscle contractility without elevating energy expenditure (31). In addition, more efficient breathing may fasten liberation from mechanical ventilation (32). This may be important for patients with respiratory failure such as acute exacerbation of COPD. Of note, our previous *in vitro* work showed that the effect of levosimendan is more profound in type-1 diaphragm fibers than in type-2 fibers (12). Because a fiber type shift towards type-1 fibers is known to occur in the diaphragm of patients with COPD (8) and congestive heart failure (33), levosimendan treatment can potentially be more effective in these patients than in healthy subjects. Accordingly, the rationale for evaluating the effect
of levosimendan on respiratory muscle function in patients with respiratory failure appears highly appropriate.

**Experimental model**

The contractile performance of the diaphragm was evaluated using magnetic stimulation and maximal inspiratory efforts. Magnetic stimulation has a clear benefit over voluntary maneuvers because it is effort independent and highly reproducible (34). Values for $P_{di_{tw}}$ and $P_{di_{max}}$ reported in our study are similar as reported by other groups for healthy subjects (20, 35). Due to coincidence, baseline $P_{di_{tw}}$ as well as CMAP$di\_amplitude$ were lower (though insignificant) in the levosimendan group (Table 1), suggesting these subjects were more difficult to stimulate. Correcting for CMAP$di\_amplitude$ abolished the baseline difference between the groups (data not shown).

To further evaluate the function of the diaphragm we measured neuro-mechanical efficiency of the diaphragm ($P_{di}/EMG_{di}$) during inspiratory loaded breathing. We found that diaphragm neuro-mechanical efficiency at 40% of $P_{di_{max}}$ was constant over time and that $CF_{di}$ decreased over time during loaded breathing, which are in line with previous data from Sinderby (36). We also found that, despite evidence of twitch force loss, the diaphragm neuro-mechanical efficiency was not reduced in the placebo group during the second loaded breathing task. Thus, the reductions in maximal contractility were not reflected in the neuro-mechanical efficiency during submaximal contractions. Regardless of the reason for this inconsistency, the double-blinded randomized design of the study should ensure that the finding of improved neuro-mechanical efficiency - only in the levosimendan group - was not due to bias, carry over effects, or other subjective influences.

For an accurate physiological measurement of EMG$di$ and CF$di$ during voluntary breathing it is necessary to control for changes in muscle-to-electrode distance, electrode positioning with respect to the muscle fiber direction and location, influence of cross-talk from other muscles (including the heart and the esophagus) and electrode movement-induced artifacts. The technology used to measure and process the EMG$di$ and CF$di$ in our study minimizes all these influences (37, 38).

It could be postulated that improved contractile function of the diaphragm resulted from cardiac inotropic effects of levosimendan. However, subjects in the present study did not have a medical history of cardiac disease and the effect of levosimendan on cardiac output was modest. Development of $P_{di_{tw}}$ occurs through a sharp anaerobic maneuver of the diaphragm; therefore it is unlikely that an improved cardiac output (and oxygen delivery) would explain the restored diaphragm contractility with levosimendan. Moreover, levosimendan improved efficiency already
during unloaded breathing and immediately at the start of the loading task, which is unlikely the result of improved oxygen delivery to the diaphragm.

The dose of levosimendan used in the current study was derived from earlier studies in healthy subjects, demonstrating limited side effects (39). This is in line with our study. Future studies should evaluate the effects of lower doses of levosimendan on respiratory muscle function in humans.

In conclusion, the present study demonstrates that the calcium sensitizer levosimendan improves both contractile function and neuro-mechanical efficiency of the human diaphragm. These findings suggest a new therapeutic approach for patients with acute respiratory muscle dysfunction.

**Acknowledgment**

The authors thank Dr. M. Kivikko and Prof. P. Rytila from Orion Pharma, Espoo, Finland, for providing levosimendan and placebo; N. Comtois from the Department of Medicine, Division of Critical Care Medicine, St. Michael’s Hospital, University of Toronto, Canada and Dr. H. van Dijk from the Department of Neurology, Radboud University Nijmegen Medical Centre, for technical assistance; J. van den Brule from the Department of Critical Care Medicine, Radboud University Nijmegen Medical Centre for performing the transthoracic echocardiography; and the research nurses from the Department of Critical Care Medicine, Radboud University Nijmegen Medical Centre for assistance during the experiments.
Levosimendan improves human diaphragm function

References


Levosimendan improves human diaphragm function


Supplemental material

Supplemental methods
Randomization and masking
Healthy subjects were randomly assigned to receive either levosimendan or placebo (ratio 1:1). The randomization code list, with a block size of six, was generated by an independent investigator of the Radboud University Nijmegen Medical Centre. The investigator and subject were masked to treatment. Both the active compound (levosimendan) and the placebo were similar in appearance (yellow solution) and could not be distinguished from one another. Levosimendan and placebo were labelled and stored by the department of Clinical Pharmacology of the Radboud University Nijmegen Medical Centre.

Esophageal catheter and its positioning
Diaphragm electromyogram (EMGdi) was measured via an esophageal catheter (NeuroVent Research Inc, Toronto, Canada) with nine stainless steel wire electrode rings placed 16 mm apart on 8 French polyurethane triple lumen tubing (2.5 mm diameter) with the tenth ring, the ground, placed 4 cm above the most proximal ring, creating an array of eight sequential electrode pairs. Two 4 cm long, 1.5 cm diameter polyurethane balloons were mounted 2.75 cm below the most distal ring and 1 cm above the ground ring for the measurement of gastric (Pga) and esophageal (Pes) pressures, respectively. The catheter was passed through the nose, swallowed, and the electrodes were positioned near the gastroesophageal junction (1, 2). The balloons were connected to two differential pressure transducers (range ± 50 kPa, Freescale, Tempe, AR) via two PVC tubes (0.75 mm diameter) inserted in each pressure lumen. The balloons were filled with 1.5 ml of air in order to distend the walls of the balloons evenly. Following this, air was withdrawn until the desired amount (0.3 – 0.4 ml) remained in the balloon.

Magnetic stimulation and maximal inspiratory effort
Cervical magnetic stimulation of the phrenic nerve was performed using a 90 mm circular coil (P/N 9784-00) powered by a Magstim 200² stimulator (Magstim Company Ltd, Whitland, UK). While seated with their neck flexed, a closed mouth and wearing a nose clip, magnetic stimulation was performed at functional residual capacity (FRC) with unbound abdomen. Stimulation position was determined and marked according to Similowski and colleagues (3). Subsequently, the increase in twitch Pdi (Pdi_{tw}) and compound muscle action potential of the diaphragm (CMAPdi) were measured at 100% Magstim output as the mean values of three twitches at least 30 seconds apart.
Pressure generating capacity of the diaphragm can be assessed by different techniques, including $P_{\text{di,sniff}}$ and maximal inspiratory effort against occluded airway. The latter technique was selected as this value was needed to calculate the load during the loading task (i.e. 40% maximal $P_{\text{di}}$).

**Data acquisition**

EMGdi signals were amplified and digitized (Porti 16, 22 bits, 71.5 nV/least significant bit, TMSi; The Netherlands) at a sampling frequency of 2 kHz. Pressure signals were digitized (Porti 16, 22 bits, 1.4 µV/least significant bit, TMSi; The Netherlands) at a sampling frequency of 100 Hz. Data were stored and buffered on a hard disk for offline analysis in Matlab (R2009b, The Mathworks, Natick, MA) and to provide visual feedback.

**Signal processing**

The segments of EMGdi used in the analysis were automatically selected between the electrocardiogram R-R intervals to avoid contamination of the QRS complex. The relative position of the centre of the electrical active region of the contracting diaphragm with respect to the electrode array was determined (1, 2). The “double subtraction technique” was then applied to enhance EMGdi signal quality (2). The double subtracted signal was converted from the time domain into the frequency domain by fast Fourier transform. Signal quality was evaluated from the power spectrum according to Sinderby and coworkers (4). For those signals deemed to be of acceptable quality, we calculated EMGdi (as the root mean square [RMS]) from the time domain and center frequency of the diaphragm ($C_{\text{di}}$) from the EMGdi power spectrum. During loading, EMGdi and $C_{\text{di}}$ were calculated as means per 10 second isometric contraction.

Compound muscle action potentials of the diaphragm (CMAPdi) were recorded from the electrode pair with the largest amplitude. Visual inspection for each individual CMAPdi was performed to avoid contamination of the QRS complex or motion artifacts. CMAPdi amplitude was calculated as the maximal displacement from negative to positive peak value. Mean values of three measurements were calculated.

Twitch transdiaphragmatic pressure ($P_{\text{di, tw}}$) and maximum transdiaphragmatic pressure ($P_{\text{di, max}}$) were calculated as the displacement from baseline to peak value. Mean values of three measurements were calculated. During loading, voluntary $P_{\text{di}}$ was calculated as means per 10 second isometric contraction.
Relation between center frequency and muscle fiber conduction velocity

As reported previously (5), there are strong reasons to assume that changes in CFdi, as measured in the present study, represent changes in muscle fiber conduction velocity. The muscle fiber conduction velocity depends on the cable properties of the fiber, which remain relatively stable during muscle contractions (6), and the membrane excitability (7, 8). The latter depends on ion gradients across the membrane generating the driving electric force and the properties of the proteins making up the gating ion channels. If the electrolyte imbalance across the sarcolemma leads up to a slight depolarization of the membrane, inactivation of the voltage-dependent sodium channels occurs, reducing membrane excitability and slowing propagation of the action potentials (7-9). Proportionality between APCV and the power spectrum frequency shift has been theoretically described in detail (1, 10), and more recently described in a review as a method to estimate muscle fiber conduction velocity (11).

Supplemental Figure 1. Center frequency of the diaphragm electromyogram (CFdi) at one, three, six, and nine minutes into the first and second loading task in the placebo (A) and levosimendan (B) group. CFdi decreased over time (P<0.001) in both loading tasks in the placebo and levosimendan group. Administration of levosimendan resulted in a downward baseline shift in CFdi (P<0.05), whereas administration of placebo had no effect on CFdi. Data are presented as mean ± SEM (placebo n=13, levosimendan n=14).
Supplemental references


Chapter 10

Effects of experimental human endotoxemia on diaphragm function

Jonne Doorduin, Jenneke Leentjens, Matthijs Kox, Hieronymus W.H. van Hees, Johannes G. van der Hoeven, Peter Pickkers, Leo M.A. Heunks

Abstract

Introduction: Systemic inflammation is a well-known risk factor for respiratory muscle weakness. Studies utilizing animal models of inflammation have shown that endotoxin administration induces diaphragm dysfunction. However, the effects of in vivo endotoxin administration on diaphragm function in humans have not been studied. Our aim was to evaluate diaphragm function in a model of systemic inflammation in healthy subjects.

Methods: Two groups of 12 male volunteers received an intravenous bolus of 2 ng/kg of E. coli lipopolysaccharide (LPS) and were monitored until 8 hours after LPS administration. In the first group twitch transdiaphragmatic pressure (Pdi\textsubscript{tw}) and compound muscle action potential of the diaphragm (CMAP\textsubscript{di}) were measured. In addition, plasma levels of cytokines, heart rate and arterial blood pressure were measured. In the second group, catecholamines as well as respiratory rate and blood gas values were measured. Diaphragm ultrasonography was performed in four subjects with severe shivering.

Results: LPS administration resulted in flu-like symptoms, hemodynamic alterations, and increased plasma levels of cytokines. Pdi\textsubscript{tw} increased after LPS administration from 31.2±2.0 cmH\textsubscript{2}O (baseline) to 38.8±2.0 cmH\textsubscript{2}O (t=1 hr) and 35.4±2.0 cmH\textsubscript{2}O (t=1.5 hr). There was no correlation between cytokine plasma levels and Pdi\textsubscript{tw}. We found a trend towards a gradual decrease in CMAP\textsubscript{di} from 0.78±0.07 mV (baseline) to 0.58±0.05 mV (t = 2 hr ). Respiratory rate increased after LPS administration from 16.8±0.5 breaths/min (baseline) to 20.3±0.6 (t=4 hr) with a resulting decrease in PaCO\textsubscript{2} of 0.5±0.1 kPa. Plasma levels of epinephrine peaked at t=1.5 hr with an increase of 1.3±0.3 nmol/L from baseline. Rapid diaphragm contractions consistent with shivering were observed.

Conclusions: This study shows that, in contrast to diaphragm dysfunction observed in animal models of inflammation, in vivo diaphragm contractility is augmented in the early phase following low dose endotoxin administration in humans.
Introduction

Respiratory muscle weakness frequently occurs in the critically ill and is associated with prolonged weaning from mechanical ventilation and increased mortality (1, 2). The pathophysiology of intensive care unit-acquired respiratory muscle weakness is complex, but inflammation is a well-known risk factor (2-4). Moreover, sepsis is a major independent risk factor for diaphragm dysfunction on intensive care unit admission (2). Several studies utilizing different animal models have shown that systemic inflammation reduces force-generating capacity of the diaphragm (3, 5). Cytokines are proposed to play an important role in affecting contractile function during inflammation. For example, tumor necrosis factor (TNF)-α has been shown to mediate the detrimental effects of endotoxin administration on contractile function of the diaphragm (6). In addition, data from our lab revealed that plasma from septic shock patients induces atrophy in cultured skeletal muscle cells within 24 hours with a prominent role for interleukin (IL)-6 (7).

Experimental human endotoxemia is a well-characterized, validated model of systemic inflammation in healthy subjects, that, among other effects, results in subclinical end-organ dysfunction (8-12). Up till now, the effects of endotoxin administration on non-volitional respiratory muscle function in humans have not been studied. Such a study could provide valuable information as to whether human endotoxemia can serve as a model for early inflammation-induced diaphragm dysfunction. If true, this would be a suitable model to test therapeutic agents aimed to improve respiratory muscle function in critically ill patients.

The objective of the present explorative study was to investigate changes in the function of the diaphragm in vivo during experimental human endotoxemia. We hypothesized that administration of endotoxin impairs diaphragm function.

Materials and methods

Ethics statement
All participants gave written informed consent prior to the study. This study was performed in subgroups of 2 larger endotoxin trials (13). Both studies were approved by the ethical committee of the Radboud University Medical Center (NL36068.091.11 and NL42337.091.12) and in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments.
Study population
In the first group of 12 subjects, we determined diaphragm function during experimental endotoxemia. As a consequence of the results obtained in that study, respiratory rate, blood gas values and catecholamines were determined in 12 additional subjects that underwent experimental endotoxemia in a later trial. Diaphragm function was not evaluated in these subjects. In both groups, no intervention besides endotoxin administration was applied.

Esophageal catheter
Prior to LPS administration, subjects were instrumented with an 8 French polyurethane esophageal catheter (NeuroVent Research Inc, Toronto, Canada). This catheter was equipped with two 4 cm long, 1.5 cm diameter polyurethane balloons for the measurement of gastric (Pga) and esophageal (Pes) pressure, and 9 electrodes to record diaphragm electromyography (EMGdi). The catheter was passed through the nose and swallowed. Proper balloon position was confirmed in all subjects by observing cardiac oscillations and esophageal spasms in Pes, whereas these artefacts were not visible in Pga. With the correct position of the catheter, the esophageal balloon was located in the lower third of the esophagus and the gastric balloon in the stomach. The balloons were connected to two differential pressure transducers (range ± 50 kPa, Freescale, Tempe, AR) via two PVC tubes inserted in each pressure lumen. The balloons were filled with 1.5 ml of air in order to distend the walls of the balloons evenly. Following this, air was withdrawn until the desired amount (0.3-0.4 ml) remained in the balloon. Transdiaphragmatic pressure was calculated as gastric pressure minus esophageal pressure.

Experimental human endotoxemia
The protocol for experimental human endotoxemia was identical in both groups. Subjects were screened by physical examination, electrocardiography, and routine laboratory values. Exclusion criteria were: febrile illness during the 2 weeks before the endotoxemia experiment, taking any prescription medication, or participation in a previous trial where LPS was administered.

After local anesthesia, the radial artery of the non-dominant arm was cannulated for blood pressure monitoring and blood sampling. A second cannula was placed in a deep antecubital vein for prehydration. Subjects received an intravenous bolus of 2 ng/kg of US Standard Reference E. coli endotoxin (Escherichia coli O:113, Clinical Center Reference Endotoxin, National Institute of Health, Bethesda, MD [LPS]) in 1 minute at t=0 hr. Concentrations of TNF-α, IL-6, IL-10 and IL-1Ra in plasma were analyzed using a simultaneous Luminex assay (Milliplex; Millipore, Billerica, MA, USA)
at t=0 hr (baseline), and t=1, 1.5, 2, 4, and 8 hr after LPS administration. Respiratory rate was recorded and blood gas parameters were analyzed. Furthermore, blood was collected into chilled lithium-heparin tubes and was immediately placed on ice and centrifuged at 2000g for 10 minutes at 4°C after which plasma was stored at -80°C until analysis. Plasma norepinephrine, epinephrine, and dopamine concentrations were measured using routine analysis methods also used for patient samples (high-performance liquid chromatography with fluorometric detection, as described previously (14)) .

**Cervical magnetic stimulation**

To evaluate diaphragm contractility and excitability, twitch Pdi (Pdi\(_{tw}\)) and compound muscle action potential of the diaphragm (CMAP\(_{di}\)), respectively, were assessed by cervical magnetic stimulation of the phrenic nerves. Magnetic stimulation of the phrenic nerve was performed using a 90 mm circular coil (P/N 9784-00) powered by a Magstim 200\(^2\) stimulator (Magstim Company Ltd, Whitland, UK). Magnetic stimulation makes use of electromagnetic induction. The magnetic field produced by the circular coil penetrates the skin and induces a secondary current in the tissue which causes depolarisation of the phrenic nerve. Consequently, the action potentials travels along the phrenic nerve and elicits a short and synchronized response (twitch) in the diaphragm. To obtain a reproducible response we used the following protocol. First, cervical stimulation position was determined and marked according to Similowski and colleagues (15). Briefly, subject were seated in an upright position with the neck flexed slightly, a closed mouth and wearing a nose clip. The area for optimal phrenic nerve stimulation was located by moving the magnetic coil around the spinous process of C7. Thereby stimulating the roots of C3-C5 from which the phrenic nerve originates. Magnetic stimulation was performed at the end-expiratory phase (by observing the subject, and pressure waveforms) with unbound abdomen. The position associated with the response of greatest amplitude at 70% stimulation intensity was carefully marked on the skin to ensure consistency in stimulation. Second, stimulation was performed at 100% Magstim output at least 30 seconds apart to avoid diaphragm potentiation. Stimulation was performed at t=0 hr (baseline), and t=1, 1.5, 2, 4, and 8 hr following LPS administration.

**Data acquisition and analysis**

Pressure signals were digitized (Porti 16, 22 bits, 1.4 µV/least significant bit, TMSi; the Netherlands) at a sampling frequency of 2 kHz. EMG\(_{di}\) signals were amplified and digitized (Porti 16, 22 bits, 71.5 nV/least significant bit, TMSi; the Netherlands) at a sampling frequency of 2 kHz. Data were stored on a hard disk for off-line analyses
in Matlab (R2012b, The Mathworks, Natick, MA). Recordings were visually inspected to discard recordings with contamination of the QRS complex, esophageal spasms or motion artefacts. CMAPdi was recorded from the electrode with the largest amplitude and calculated as the peak-to-peak displacement. Pdi\textsubscript{tw} was calculated as the peak difference from baseline. See Figure 1 for examples of CMAPdi and Pdi\textsubscript{tw}. Mean values of three to five measurements were calculated.

![Figure 1](image.png)

**Figure 1.** Example of CMAPdi and Pdi\textsubscript{tw} following magnetic phrenic nerve stimulation. Magnetic stimulation of the phrenic nerves elicits a compound muscle action potential of the diaphragm (a) with a subsequent increase in transdiaphragmatic pressure (b). Gray lines represent 5 individual stimulations and the black bold line is the average response. Note the difference in time scale between a and b. Abbreviations: CMAPdi = compound muscle action potential of the diaphragm; Pdi\textsubscript{tw} = twitch transdiaphragmatic pressure.

**Diaphragm ultrasonography**

In subjects with severe shivering, and a good ultrasonographic view of the diaphragm, diaphragm ultrasonography was performed to examine shivering of the diaphragm at 1 hour following LPS administration. Ultrasonography was performed using two windows: (1) a linear array transducer in the zone of apposition (16, 17), an echogenic layer bordered by pleural and peritoneal membranes; and (2) a phased array transducer positioned subcostal or low intercostal between the midclavicular and mid-axillary lines (18).

**Statistical analysis**

To evaluate diaphragm function, plasma cytokines, plasma levels of catecholamines, respiratory rate, and blood gas values during experimental endotoxemia we performed repeated measures one-way ANOVA for normally distributed data, or the Friedman test as its non-parametric equivalent. Repeated measures testing
Effects of experimental human endotoxemia on diaphragm function

was not possible for Pdi_tw and CMAPdi due to missing values at different time points. Therefore, we used the change from baseline to correct for inter-individual differences and performed one-way ANOVA for normally distributed data, or the Friedman test as its non-parametric equivalent. Post-hoc analysis was performed with Tukey’s test or Dunn’s test, as its non-parametric equivalent, to correct for multiple comparisons. Spearman’s correlation was performed to test associations between plasma cytokines and diaphragm function. A level of $p<0.05$ (two-tailed) was considered as statistically significant. The normality of the distribution of the different variables was evaluated with the D’Agostino & Pearson test. Data are described as mean±standard error or median [interquartile range] depending on the distribution of the data. Statistical analysis was performed with Graphpad Prism 5 (Graphpad software, San Diego, CA, USA).

Results

Baseline characteristics of the subjects are presented in Table 1. Experimental endotoxemia resulted in the expected flu-like symptoms in all subjects. Symptoms started 45 to 60 min after LPS administration with a brief episode of shivering, followed by varying degrees of headache, nausea, general malaise and myalgia. Symptoms completely subsided after 5-6 hrs in all volunteers. Due to the described symptoms, magnetic phrenic nerve stimulation was not feasible in every subject at every time point. Furthermore, in two subjects the esophageal catheter was removed after 1 and 1.5 hr following LPS administration because of unacceptable discomfort. The number of subjects per time point is given in the legend of Figure 2.

There was an expected increase in plasma concentrations of cytokines. Pro-inflammatory cytokines TNF-α and IL-6 peaked at t = 1.5 and 2 hr, respectively (Figure 2a-b), while anti-inflammatory cytokines IL-10 and IL-1Ra peaked at t = 2 and 4 hr (Figure 2b). Pdi_tw increased from baseline (31.2 ± 2.0 cmH_{2}O) after LPS administration (Figure 2c) with a significant peak at t = 1 hr. Note that only six and eleven patients could be evaluated 1 and 1.5 hour after LPS administration, respectively, due to severe malaise/shivering. Nevertheless, increases in Pdi_tw at t = 1 and t = 1.5 hr from baseline were robust and significant in this set of subjects (Figure 3a-b). No significant changes were found in CMAPdi over time (Figure 2d), however a decreasing trend was observed after LPS administration, reaching its nadir after 2 hours. Indeed, at t = 2 hr CMAPdi decreased in almost all analyzed subjects (Figure 3c). LPS administration resulted in an increase in heart rate and decrease in mean arterial pressure (Figure 2d).
Correlations between Pdi\textsubscript{tw}, CMAP\textsubscript{di} and cytokine levels were explored at different time points (see supplemental Figure 1 and supplemental Figure 2). No consistent correlations between Pdi\textsubscript{tw} or CMAP\textsubscript{di} and cytokine levels were found.

In addition, we found a minor, but statistically significant, increase in breathing frequency following LPS administration (Figure 4a). Blood gas analysis revealed an increase in pH due to a decrease in PaCO\textsubscript{2} (Figure 4b). PaO\textsubscript{2} did not change during endotoxemia (Figure 4b). Plasma epinephrine concentrations increased after LPS administration with a sharp peak at t = 1.5 hr, whereas plasma dopamine and norepinephrine concentrations were only slightly altered and remained within the reference range throughout the endotoxemia experiment (Figure 4d).

Diaphragm ultrasonography was performed in selected subjects with severe shivering (n=4). Rapid diaphragm contractions consistent with shivering were observed in these subjects.

**Figure 2.** Cytokine levels, diaphragm function and hemodynamic measurements. Panels a and b depict concentrations of pro- (TNF-\(\alpha\), IL-6, and IL-10) and anti-inflammatory (IL-1Ra) cytokines after LPS administration (n=12). Panels c and d show Pdi\textsubscript{tw} and CMAP\textsubscript{di}, respectively, after LPS administration. In panels c and d the number of subjects are n=12, 6, 11, 9, 10 and 10 for time points t=0, 1.5, 2, 4 and 8 hr, respectively. Panel e depicts HR and MAP after LPS administration (n=12). Data are presented as median [interquartile range] (panels a, b and c) and mean ± standard error (panel d and e). The p-value in the panels represent result of the repeated measures one-way ANOVA for normally distributed data, or the Friedman test as its non-parametric equivalent. Asterisks represent significant changes per time point from baseline as evaluated by post-hoc analysis with Tukey’s test or Dunn’s test, as its non-parametric equivalent. Abbreviations: CMAP\textsubscript{di} = compound muscle action potential of the diaphragm; HR = heart rate; MAP = mean arterial blood pressure; Pdi\textsubscript{tw} = twitch transdiaphragmatic pressure.
Table 1. Subject demographic characteristics

<table>
<thead>
<tr>
<th></th>
<th>Group 1 (original experiment, n=12)</th>
<th>Group 2 (additional experiments, n=12)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex (M/F)</td>
<td>(12/0)</td>
<td>(12/0)</td>
<td></td>
</tr>
<tr>
<td>Age, yrs</td>
<td>21 ± 0.4</td>
<td>23 ± 0.8</td>
<td>0.15</td>
</tr>
<tr>
<td>Height, cm</td>
<td>183 ± 3</td>
<td>184 ± 1</td>
<td>0.69</td>
</tr>
<tr>
<td>Weight, kg</td>
<td>79 ± 2.7</td>
<td>79 ± 2.1</td>
<td>0.88</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>23.6 ± 0.6</td>
<td>22.9 ± 0.6</td>
<td>0.49</td>
</tr>
<tr>
<td>HR, beats/min</td>
<td>68 ± 2.5</td>
<td>61 ± 2.8</td>
<td>0.09</td>
</tr>
<tr>
<td>MAP, mmHg</td>
<td>95 ± 2.5</td>
<td>94 ± 2.4</td>
<td>0.87</td>
</tr>
</tbody>
</table>

Parameters measured during screening visit. Data are presented as means ± standard error. Abbreviations: BMI = body mass index; HR = heart rate; MAP = mean arterial blood pressure. P-values calculated using unpaired t test.

Figure 3. Increase in Pdi\textsubscript{tw} at t=1 and t=1.5 hr and decrease in CMAPdi at t= 2 hr Panels a and b depict individual increases in Pdi\textsubscript{tw} from baseline (t=0 hr) to t=1 and t=1.5, respectively. Note that only six and eleven patients could be evaluated 1 and 1.5 hour after LPS administration, respectively, due to severe malaise/shivering. Panel c shows individual decreases in CMAPdi (n=8) from baseline (t=0 hr) to t=2 hr. Abbreviations: CMAPdi = compound muscle action potential of the diaphragm; Pdi\textsubscript{tw} = twitch transdiaphragmatic pressure.
Figure 4. Respiratory parameters and catecholamine concentrations. Panel a depicts breathing frequency after LPS administration (n=12). Panels b and c depicts blood gas values (pH, PaCO₂ and PaO₂) after LPS administration (n=12). Panel d depicts plasma catecholamine concentration (dopamine, epinephrine, nor-epinephrine) after LPS administration (n=12). Data are presented as median [interquartile range]. The p-value in the panels represent result of the Friedman test. Asterisks represent significant changes per time point from baseline as evaluated by post-hoc analysis with Dunn’s test. P-values calculated using Friedman test for epinephrine, norepinephrine, and dopamine are p < 0.0001, p = 0.0029, and p = 0.0054, respectively.
Discussion

In the present study we used a standardized, well-validated human model to investigate the effects of systemic inflammation on non-volitional diaphragm function in vivo. In contrast to our hypothesis, experimental endotoxemia did not attenuate but temporarily enhanced diaphragm contractility, despite a trend towards gradual decrease in diaphragm muscle fiber excitability.

Experimental model

The contractile performance of the diaphragm during experimental human endotoxemia was evaluated using magnetic phrenic nerve stimulation. Magnetic stimulation has a clear benefit over voluntary manoeuvres because it is independent of subject motivation, cooperation, and effort. The baseline values for Pdi in our study are similar to those reported previously (19).

To our knowledge this is the first study that describes diaphragm contractility following LPS administration in humans. In apparent contrast with our hypothesis and previous findings in animal models and sepsis patients, diaphragm force did not decrease, but increased during systemic inflammation induced by experimental endotoxemia. Animal studies describe a significant impairment of diaphragmatic force, becoming evident 4 hours after endotoxin administration (3, 6). It should be noted that the dose of LPS administered in our study is much lower (2 ng/kg) compared with animal experiments which typically utilize (sub)lethal LPS dosages in the order of milligrams per kilogram bodyweight (6). It is very well possible that the dose required to induce diaphragm dysfunction might be considerably higher. In critically ill patients, sepsis was recently identified as a major independent risk factor for diaphragm dysfunction on intensive care unit admission (2). In the present study, LPS administration clearly elicited sepsis-like responses such as flu like symptoms and hemodynamic alterations as well as elevated cytokine levels. Furthermore, experimental human endotoxemia has been shown to result in, although mild, cardiac (8), vascular (9, 10), renal (11), and coagulatory (12) dysfunction. Nevertheless, this model does not accurately represents all organ dysfunction as observed in sepsis patients (20, 21). There are many discrepancies (20, 21), including the fact that the inflammatory response is very short-lived. Previously, the effects of experimental human endotoxemia on quadriceps muscle function have been studied (22). A reduction in voluntary muscle contractile force was found (22), which likely resulted from poor volition caused by the malaise associated with endotoxemia, as force generation upon stimulation was not affected by endotoxin. Interestingly, using approximately the same dose of
endotoxin (4 ng/kg), Suffredini and colleagues found depression of left ventricular function during endotoxemia in healthy subjects (8). This might indicate that cardiac muscle is more susceptible to endotoxin-induced inflammation than peripheral skeletal muscle or the diaphragm.

In contrast to increased diaphragm contractility, we observed a trend towards gradual decrease in diaphragm membrane excitability during endotoxemia. This is consistent with animal studies, where it was shown that a sublethal dose of TNF-α reduces CMAPdi in response to phrenic nerve stimulation (23), and that contractile dysfunction was found to be associated with a decreased diaphragmatic resting membrane potential in endotoxic shock (24). The latter phenomenon could impair action potential generation resulting in failure of neuromuscular transmission. Nevertheless, in our study diaphragm contractility was not compromised, suggesting that other sites in the excitation-contraction coupling compensated for the minor decrease in CMAPdi.

Taken together, experimental human endotoxemia does not represent an appropriate in vivo model for systemic inflammation-induced diaphragm dysfunction and can therefore not be used to test new therapeutic agents aimed to improve diaphragm function in critically ill patients.

Mechanisms of action
The mechanism behind the increase in Pdi_tw after endotoxin administration is unknown. We reasoned that an increased Pdi_tw could be the result of activation of beta receptors on respiratory muscle. Indeed, β2-adrenoreceptor agonist treatment increases calcium release from the sarcoplasmatic reticulum and improve diaphragm contractility in rats (25, 26). In our study endogenous epinephrine levels were higher compared with baseline when Pdi_tw increased (Figure 2d). However, Pdi_tw peaked before the peak in plasma epinephrine levels at t= 1.5 hr.

In the diaphragm the twitch response is affected by prior voluntary efforts, a phenomenon referred to as twitch potentiation. Therefore, potentiation of the diaphragm might cause an increase in Pdi_tw. For example, five seconds of 25% of maximal voluntary diaphragm contractions may already potentiate the diaphragm and result in higher twitch pressures (27). Respiratory rate increased more than 2 hours after endotoxin administration in our model, which is in accordance with previously published data (28), but at that time-point Pdi_tw already decreased, which makes potentiation of the diaphragm due to increased respiratory activity an unlikely explanation for the increase in Pdi_tw at 1 hour (Figure 2c and 4a). An alternative hypothesis is a potentiating effect of shivering thermogenesis of the diaphragm. Experimental studies have shown that phosphorylation of the skeletal
muscle ryanodine receptor 1 (a calcium release channel) occurs during shivering thermogenesis (29). Interestingly, phosphorylation of calcium release channels may play a role in twitch potentiation (30, 31). Most subjects in our study experienced a period of shivering starting 1 hour after LPS administration, coinciding with the increased $P_{di}^{tw}$ levels. Indeed, diaphragm ultrasonography 1 hour after LPS administration revealed shivering of the diaphragm. Thus, shivering thermogenesis in the early phase of endotoxemia might have had a potentiating effect on the diaphragm.

In future research, this hypothesis could be partly tested by performing voluntary potentiation. For voluntary potentiation, phrenic nerve stimulation is performed after the subject makes a maximal voluntary contraction. If no further potentiation occurs it is possible that previous potentiation (due to endotoxemia or hyperventilation) took place. In addition, measurement of work of breathing, pressure-time product, and neuromechanical coupling during human endotoxemia may contribute to elucidate our results.

**Study limitations**

The human endotoxemia model used in our study, and most animal endotoxemia models, relate to the early changes observed with systemic inflammation and is thus a model of short-lived systemic inflammation and not a model of sepsis. This should be kept in mind while interpreting our data, since different mechanism may affect muscle function during the early phase of systemic inflammation compared to the long term effects of sepsis (32).

With increasing lung volume, there is a linear decrease in $P_{di}^{tw}$ (33). Therefore, it is of importance to control for lung volume when assessing diaphragm contractility. We were not able to measure lung volumes using spirometry prior to stimulation. However, we aimed to perform all stimulations at functional residual capacity. Functional residual capacity was ensured by performing magnetic stimulation at the end-expiratory phase by observing the subject, and Pes and Pga waveforms.

Ideally, all measurements should have been performed in one human endotoxemia study. However, respiratory rate, blood gas values and catecholamines were determined in a different group than diaphragm function. This was a consequence of scientific reasoning, since the results of the first experiment were in contradiction to our hypothesis and we wanted to further explore possible mechanisms behind the observed effects. The goal of the second experiment was to find the mechanism of action for increased diaphragm function. Due to the invasive nature of the diaphragm function measurements we chose not to perform these in the second experiment. Nonetheless, we used the same well-characterized and validated experimental
model of systemic inflammation with a virtually identical protocol, and subject characteristics were comparable as well. Therefore, we assume that performing the study in two separate experiments has not confounded our results.

Conclusions

In conclusion, our study shows that, in contrast to animal experiments, \textit{in vivo} diaphragm contractility is augmented in the first hours following low dose endotoxin administration in humans. Therefore experimental human endotoxemia does not represent an appropriate \textit{in vivo} model for systemic inflammation-induced diaphragm dysfunction.
References


Effects of experimental human endotoxemia on diaphragm function
Supplemental Figure 1. Correlations between twitch transdiaphragmatic pressure (Pdi_tw) and cytokine levels at different time points. HR, hour.
Supplemental Figure 2. Correlations between compound muscle action potential of the diaphragm (CMAPdi) and cytokine levels at t = 2 hour.
Chapter 11

Summary, general discussion and future perspectives
Chapter 11

Summary

In the last decades, evidence has accumulated that diaphragm dysfunction frequently develops in critically ill patients and contributes to prolonged mechanical ventilation. However, monitoring of diaphragm function during mechanical ventilation and weaning has received little attention. In fact, most clinicians do not monitor diaphragm function in critically ill patients at all. In addition, there are limited interventions available that target the diaphragm. In this thesis, techniques have been investigated to monitor diaphragm function, and preventive and therapeutic interventions to optimize diaphragm function.

The first part of this thesis focuses on monitoring of diaphragm function in critically ill patients. In chapter 2, a clinical perspective, we discussed the pros and cons of the available techniques to monitor respiratory muscle function. These techniques include easy to learn and non-invasive tools, such as maximal inspiratory pressure and diaphragm ultrasound. But also complex and sophisticated measurements, such as phrenic nerve stimulation, electrical activity of the diaphragm (EAdi), and transdiaphragmatic pressure (Pdi). Some tools have limited value (e.g. fluoroscopy, occlusion pressure in the first 100ms) or are not suitable for routine monitoring (e.g. phrenic nerve stimulation). However, promising tools that can be used today or in the near future for monitoring respiratory muscle function are recordings of Pdi and EAdi, and diaphragm ultrasound. Although there is no literature yet that directly demonstrates improved outcome with close monitoring (and action) of the respiratory muscles, in our opinion monitoring the respiratory muscles should be as much part of the routine as monitoring any other vital organ function.

Weaning from mechanical ventilation is an important phase for critically ill patients. The transition from positive inspiratory pressure during mechanical ventilation to negative pleural pressure during spontaneous breathing challenges the patient's physiological reserve. When there is an imbalance between the patient's ventilatory needs and the capacity of the neural, cardiovascular and respiratory systems, weaning fails. In chapter 3 we reviewed the pathophysiology of difficult and prolonged weaning and the techniques that are most helpful in the diagnostic work-up. Impaired respiratory mechanics, cardiac dysfunction, respiratory muscle dysfunction, cognitive dysfunction and metabolic disorders are recognized causes for weaning failure. Currently available research has mostly focused on respiratory muscle dysfunction and cardiac dysfunction as a cause for weaning failure. Understanding the complex pathophysiology of weaning failure in combination with a systematic diagnostic approach allows identification of the primary cause of weaning failure.
In chapter 4 we performed detailed analysis of inspiratory muscle function, expiratory muscle function and respiratory mechanics during a spontaneous breathing trial in a heterogeneous group of critically ill patients. We found that the expiratory muscles significantly contribute to respiratory muscle effort (i.e. up to 26% of total respiratory muscle effort) in patients who fail a trial of spontaneous breathing. In addition, neuromechanical efficiency of the diaphragm is 41% lower in weaning failure patients compared to patients that are successfully extubated. This confirms that impaired pressure-generating capacity of the diaphragm, regardless of its origin, plays a central role in failure to wean from mechanical ventilation.

Results from chapter 5 showed that in healthy subjects, strain and strain rate of the diaphragm, obtained by speckle tracking imaging, are highly correlated with Pdi and EAdi during inspiratory loading from zero to 60 cmH₂O. Remarkably, diaphragm thickening during inspiratory loading was not correlated with Pdi and EAdi. These findings suggest that advanced ultrasound techniques, such as speckle tracking imaging, may be used in the future to non-invasively assess diaphragmatic effort in critically ill patients.

The second part of this thesis focuses on preventive and therapeutic interventions to optimize diaphragm function. An important preventive intervention is to minimize the harmful effects of mechanical ventilation on diaphragm function, for example by optimizing patient-ventilator interaction. Neurally adjusted ventilatory assist (NAVA) is a mode of mechanical ventilation that uses the electrical activity of the diaphragm to control timing and magnitude of assist with invasive and non-invasive mechanical ventilation.

In chapter 6, patient-ventilator interaction was quantified using an automated analysis in COPD patients with different modes of noninvasive ventilation, including NAVA. We showed that NAVA synchronizes assist to the patients inspiratory effort, whereas pressure support modes of a dedicated NIV ventilator or ICU ventilator did not ensure optimal patient-ventilator interaction. In addition, wasted efforts increased exponential after timing errors between electrical activity of the diaphragm and airway pressure reached more than 20%. Thus progressive mismatch between neural effort and pneumatic timing of the ventilator is strongly associated with severe forms of patient-ventilator asynchrony. Despite these physiological effects NAVA did not improve blood gas values and patient comfort.

In ARDS patients, lung-protective ventilation is applied via controlled mechanical ventilation to allow strict control of tidal volume and plateau pressure. However, this strategy usually requires high levels of sedation and is associated with adverse events, in particular diaphragm dysfunction. In contrast, assisted modes that allow
spontaneous breathing efforts reduce the risk of diaphragm dysfunction, but may result in high tidal volumes and high transpulmonary pressures. In chapter 7, we evaluated in patients with mild to moderate ARDS how different modes of mechanical ventilation (assist-pressure control ventilation [PCV], pressure support ventilation [PSV] and NAVA) affect lung protective ventilation, breathing pattern variability, and patient ventilator-interaction. The three selected modes allowed for different degrees of ventilator control by the patient. The main finding of this study was that with increasing freedom for the patient to control the ventilator, tidal volume and transpulmonary pressure remained within the limits of lung-protective ventilation. Except for one patient with severe hypercapnic acidosis where tidal volume increased above 10 mL/kg. Breathing pattern variability and patient-ventilator interaction improved with NAVA compared to PCV and PSV. These findings suggest that there is a role for assisted mechanical ventilation, and active diaphragm contractions, in patients with mild to moderate ARDS without strong inspiratory efforts.

Based on the findings in chapter 7 we developed a ventilation strategy for ARDS patients that generate high tidal volumes after the transition from controlled mechanical ventilation to assisted ventilation. In a proof-of-concept study it was investigated whether low dose neuromuscular blockers can facilitate a lung-protective ventilation strategy without loss of diaphragm activity during assisted mechanical ventilation in lung injury patients with high tidal volumes (chapter 8). Such a strategy might allow a safe compromise between the benefits and risks of spontaneous breathing during mechanical ventilation. The results show that partial neuromuscular blocking of the diaphragm can be used to titrate tidal volume from approximately 10 ml/kg to 6 ml/kg with preservation of diaphragm activity at physiological levels. However, this strategy was associated with mild hemodynamic side effects, including tachycardia and elevated blood pressure, and development of hypercapnic acidosis. Future studies should carefully evaluate whether partial neuromuscular blockade is tolerated by patients for longer periods of time.

Previously, it was demonstrated in vitro that levosimendan enhances calcium sensitivity of permeabilized muscle fibers obtained from the human diaphragm resulting in a greater amount of force generated for the same level of cytosolic calcium. In chapter 9 we found that levosimendan also improves in vivo diaphragm function in healthy subjects. During isometric loading of the diaphragm, neuromechanical efficiency of the diaphragm improved with approximately 20%. Furthermore, levosimendan reversed the development of fatigue after diaphragmatic loading.

Systemic inflammation is a well-known risk factor for respiratory muscle weakness. In chapter 10 we aimed to develop an experimental in vivo model for diaphragm
dysfunction during systemic inflammation. In contrast to animal models of systemic inflammation, we found in healthy subjects an augmentation of *in vivo* diaphragm contractility in the early phase following low dose endotoxin administration. Therefore the human endotoxemia model does not seem suitable for *in vivo* testing of interventions to improve diaphragm function.
General discussion and future perspectives

Diaphragm dysfunction frequently develops in critically ill patients and is associated with increased morbidity and mortality. The studies performed in this thesis evaluated techniques to monitor diaphragm function, and preventive and therapeutic interventions to optimize diaphragm function.

Monitoring diaphragm function during mechanical ventilation and weaning

In clinical practice the diaphragm of critically ill patients is poorly monitored, the limited availability and knowledge of monitoring techniques may be one of the reasons. In chapter 2, we identified three promising techniques to monitor respiratory muscle function: 1) transdiaphragmatic pressure (Pdi); 2) electrical activity of the diaphragm (EAdi); and 3) diaphragm ultrasound. Indeed, these monitoring techniques have received growing attention in recent years.

Pdi is a specific measure of diaphragm function and requires a double balloon approach (gastric and esophageal balloon). Since esophageal pressure (Pes) represents global respiratory muscle function and only requires one balloon, this approach to monitor respiratory muscle functions has been described extensively (1-3). In mechanically ventilated patients with spontaneous breathing efforts, Pes monitoring can primarily be used to monitor respiratory muscle effort and patient-ventilator asynchrony. In chapter 7 and chapter 8 swings in Pes ranged from low to very high values, indicating that respiratory muscle activity ranged from insufficient to excessive. Both insufficient and excessive inspiratory muscle activity may result in diaphragm dysfunction (4-9). Monitoring Pes or Pdi during mechanical ventilation allows to titrate support to target specific respiratory muscle effort. A respiratory muscle effort between 5 and 10 cmH₂O is within a physiological range (10). During weaning, monitoring of Pes has been demonstrated to discriminate between weaning failure and weaning success (11). In chapter 4 we recorded Pes and gastric pressure (Pga) in patients during spontaneous breathing trials. In addition, we used Pes and Pga to calculate more sophisticated measures, such as intrinsic positive end-expiratory pressure, expiratory rise in gastric pressure, work of breathing and esophageal pressure-time product. From a diagnostic point of view these variables provide relevant information to determine the cause of weaning failure for an individual patient (12, 13). For routine monitoring of the respiratory muscles, however, this detailed analysis requires a too high level of expertise and monitoring of only Pes or Pdi seems more appropriate for routine clinical application.
Several studies have demonstrated that Pes monitoring can be used to detect patient-ventilator interaction (14-16). In this thesis, however, we monitored patient-ventilator interaction using EAdi, as described in the next paragraph.

Another application of Pes monitoring, in addition to respiratory muscle monitoring, is to guide lung-protective mechanical ventilation in patients with acute respiratory distress syndrome (ARDS). We showed that Pes, by calculating transpulmonary pressure, can be used to identify safe or lung injurious lung distending pressures during mechanical ventilation in ARDS patients (chapter 7-8).

Monitoring of electrical activity of the diaphragm has frequently been used to evaluate patient-ventilator interaction (17-22). Often asynchrony between the patient and the mechanical ventilator is scored manually. Instead, we evaluated patient-ventilator interaction using an automated algorithm, based on EAdi, in patients with chronic obstructive pulmonary disease (COPD) during noninvasive ventilation (chapter 6) and in ARDS patients during invasive ventilation (chapter 7). This automated analysis allows detection of asynchronies, such as wasted efforts, but also makes it easy to detect the more subtle dyssynchronies (i.e. timing errors), such as trigger delays and cycling-off errors. Dyssynchronies are easily missed by visual inspection of waveforms, but may be of clinical relevance, as we found that wasted efforts increase drastically after timing errors reach over 20% (chapter 6). An automated analysis using EAdi (or Pes) may be incorporated into mechanical ventilators to simplify monitoring of patient-ventilator interaction.

Additionally, EAdi may be used to monitor neural respiratory drive during mechanical ventilation and weaning. We found a large variety in neural respiratory drive in ARDS patients ventilated with 6 ml/kg (chapter 7). These findings suggest that adjusting ventilator assist to target low tidal volumes resulted in different levels of neural drive, probably due to a complex interaction between sedation, ventilatory mode and level of assist. Accordingly, monitoring EAdi can aid to optimize ventilation in ARDS to prevent over-assist or insufficient assist. However, the difficulty with EAdi is that the magnitude of the signal is strongly influenced by individual anatomy and this makes comparisons between patients difficult. Furthermore, there is no method to determine the optimal EAdi for an individual patient. Recently, attempts have been made to estimate inspiratory muscle effort from EAdi by using an conversion index (23) or determine the contribution of the patient to the inspiratory tidal volume (24). However, the reliability of this index is poor and therefore not yet clinical applicable to monitor respiratory muscle function (25). Future studies are required to develop a method that is able to determine the optimal EAdi for an individual patient.
The ratio between Pdi and EAdi represents neuromechanical efficiency (NME) of the diaphragm, in other words the ability of the diaphragm to translate neural respiratory input to mechanical respiratory output. A gradual decrease in NME over days may indicate the development of diaphragm weakness or fatigue, whereas an increase suggests recovery. We showed that NME discriminates between weaning failure and weaning success (chapter 4). In our study we calculated NME using a double-balloon esophageal catheter with multiple electrodes to allow continuous recording of Pdi and EAdi. Other groups have calculated NME of the inspiratory muscles as the ratio between the drop in mouth pressure during an inspiratory occlusion and electrical activity of the diaphragm (26-28). Using the latter method, NME was found to be predictive of weaning failure as well (26). In a small randomized clinical trial it was shown that NAVA improves NME whereas PSV does not (27). The latter demonstrates that monitoring of NME may be useful to detect trends in muscular efficiency of the inspiratory muscles during mechanical ventilation. However, another study demonstrated that this index nor its trend was associated with risk factors for diaphragmatic injury or clinical outcome (28). Further studies are required to determine the value of NME monitoring, and its most optimal an practical method, during mechanical ventilation and weaning.

Besides physiological measurements, imaging of the diaphragm using ultrasound is rapidly gaining popularity to monitor diaphragm function in the ICU. Diaphragm ultrasound can be used to evaluate thickness, thickening during inspiration and displacement of the diaphragm (29). Using ultrasound it has been demonstrated in critically ill patients that diaphragm thickness decreases during mechanical ventilation and that the thickening fraction of the diaphragm is related to the level of support provided by the mechanical ventilator (9, 30). However, correlations between physiological measurements, such as transdiaphragmatic pressure, and diaphragm thickening are low (31). Therefore, other ultrasound techniques are required. In healthy subjects we demonstrated that speckle tracking imaging of the diaphragm, an advanced ultrasound technique, is highly correlated with Pdi during a protocol of increased loading (chapter 5). This offers new perspectives to use diaphragm ultrasound to monitor diaphragmatic effort in critically ill patients.

This thesis demonstrates that monitoring of the diaphragm is feasible in critically ill patients. Recordings of Pdi (or Pes), EAdi and diaphragm ultrasound provide complementary information about the diaphragm. Ideally all three techniques should be available to clinicians, like blood pressure, electrocardiography and echocardiography for the heart. Nevertheless, implementing one of these techniques
in daily clinical care for ICU patients would already be a big step forward for the diaphragm. In our opinion monitoring respiratory muscles in the ICU should be as much part of the routine as monitoring any other organ function. Close monitoring of diaphragm function is a prerequisite to detect deterioration of diaphragm function in critically ill patients, but monitoring alone is useless if no actions can be undertaken to prevent the development of diaphragm dysfunction or improve its function. Therefore, potential interventions were also investigated in this thesis.

**Preventive and therapeutic interventions to optimize diaphragm function**

Currently, limited interventions are available to optimize diaphragm function in critically ill patients (32). In this thesis we evaluated novel interventions aimed at preventing diaphragm dysfunction (chapter 6-8) and improving diaphragm function (chapter 9-10).

An important factor contributing to diaphragm dysfunction in critically ill patients is mechanical ventilation, also known as ventilator-induced diaphragm dysfunction (VIDD) (33, 34). Diaphragm inactivity during mechanical ventilation is the most important pathophysiological trigger leading to VIDD, in particular during controlled mechanical ventilation (34). High levels of ventilator support during assisted mechanical ventilation, however, may also cause diaphragm dysfunction (8). Therefore, preventive measures must be taken to limit diaphragm inactivity during mechanical ventilation. Neurally adjusted ventilatory assist (NAVA) is ventilator mode that uses electrical activity of the diaphragm to control ventilator support (35). In COPD and ARDS patients, NAVA improves patient-ventilator interaction compared to pressure support ventilation (PSV) (chapter 6-7). Since poor patient-ventilator interaction is strongly associated with prolonged mechanical ventilation (36, 37), NAVA may potentially decrease the duration of mechanical ventilation and improve patient outcome. Furthermore, the proportionality between EAdi and ventilator assist prevents over-assist with NAVA, as seen with PSV (38-41). Despite these physiological benefits of NAVA, we did not find improvements in blood gas values and patient comfort with NAVA (chapter 6-7). The latter may result from the short period of time (30 min) the different ventilator modes were studied. Recently, it was demonstrated in a multicenter randomized controlled trial that NAVA is safe and feasible over a prolonged period of time but does not decrease ICU length of stay or mortality (42). This randomized trial was performed in heterogeneous group of patients with acute respiratory failure, whereas NAVA is mostly beneficial in specific patients groups, such as COPD or patients receiving high levels of support. Future
studies are required to determine if and in which patients groups NAVA prevents diaphragm dysfunction and improves patients outcome.

To limit inactivity of the diaphragm, mechanically ventilated patients should be ventilated with assisted ventilation as soon as possible. However, in patients with ARDS assisted ventilation may increase the risk of high tidal volumes and transpulmonary pressures \( (P_L) \) and subsequently aggravate lung injury, known as ventilator-induced lung injury (43). The use and timing for initiation of assisted mechanical ventilation in ARDS patients is thus subject of debate (44-47). In chapter 7, it was demonstrated that in selected patients with mild to moderate ARDS assisted mechanical ventilation (NAVA and PSV) can deliver support within the limits of lung-protective ventilation. But, there were also patients in this study where tidal volume and \( P_L \) increased to high levels during assisted mechanical ventilation. In these cases of failure of assisted ventilation clinicians will most likely switch back to controlled ventilation and/or administrate high dose sedatives. Alternatively, partial neuromuscular blockade of the diaphragm using low dose rocuronium also facilitates low tidal volumes and \( P_L \) during assisted mechanical ventilation and with preservation of diaphragm activity (chapter 8). This suggests a novel strategy to deliver both lung-protective ventilation and diaphragm-protective ventilation in ARDS patients. However, this strategy was also associated with mild hemodynamic side effects, including tachycardia and elevated blood pressure, and development of hypercapnic acidosis. To minimize hemodynamic consequences in the future, the increase in \( \text{PaCO}_2 \) could be made less rapid by titrating rocuronium more slowly. Further studies should focus on understanding the long-term consequences on diaphragm function using partial neuromuscular blocking and its feasibility under low levels of sedation.

Currently, there are no pharmacological agents available to improve diaphragm function in critically ill patients. Levosimendan is a calcium sensitizer approved to enhance cardiac contractility in patients with acute heart failure (48). Previously, is was shown that levosimendan also enhances \textit{in vitro} calcium sensitivity of permeabilized muscle fibers obtained from the human diaphragm (49). Furthermore, levosimendan reduced protein nitrosylation and markers of oxidative stress in endotoxemic rodents (50). In this thesis we demonstrated that levosimendan administration in healthy subjects improved contractile function and neuromechanical efficiency of the diaphragm during loaded breathing (chapter 9). This suggests a new therapeutic approach for patients with diaphragm dysfunction. Another troponin activator has been shown to improve calcium sensitivity permeabilized diaphragm fibers as well, but not yet in humans (51). Recently a large randomized controlled trial to
investigate whether levosimendan reduces the severity of organ dysfunction in adults with sepsis was performed (52). In apparent contrast to our findings, it was found that levosimendan was associated with a lower likelihood of successful weaning from mechanical ventilation (52). However, this study was not specifically designed to evaluate the effect of levosimendan on weaning and diaphragm function. Levosimendan was given in the acute phase of septic shock for its inotropic and anti-inflammatory effects, whereas we would propose to administer levosimendan in patients with prolonged weaning, beyond their acute phase, to specifically improve diaphragm function. Currently, we are investigating the effects of levosimendan on diaphragm function in a randomized controlled trial in critically ill patients (ClinicalTrials.gov NCT01721434).

In addition to the work presented in this thesis other interventions to counteract diaphragm dysfunction have been reported in the past years, in particular modulation of contractile activity through inspiratory muscle strength training (IMST) or diaphragm pacing. Three randomized trials have assessed the effects of IMST in mechanically ventilated critically ill patients (53-55). These trials found, with different training protocols, that IMST improves maximum inspiratory pressure and has no adverse effects. Moreover, in two trials weaning time was reduced (53, 54). Thus, IMST in critically ill patients with respiratory muscle weakness is tolerated and may facilitate weaning. Future studies are needed to determine the optimal training protocol, timing and patient selection.

Diaphragm pacing occurs through electrical stimulation of the phrenic nerve via an intrathoracic or intradiaphragmatic approach. Validated indications for implanted phrenic nerve stimulation are quadriplegia caused by high level spinal cord injury and permanent, or sleep-related central hypoventilation (56). The goal of diaphragm pacing in these patients is liberating the patients, at least during the day, from mechanical ventilation. In critically ill patients receiving mechanical ventilation, temporary diaphragm pacing via a transvenous phrenic approach may offer a solution to prevent diaphragm dysfunction or even improve diaphragm contractility. Recently, in an experimental animal study it was found that early transvenous phrenic nerve stimulation preserves diaphragm thickness and myofiber cross-sectional area in ventilated-paced pigs compared to ventilated-not paced pigs (57). The prevention of diaphragm atrophy with diaphragm pacing has also been reported in a preliminary study of three mechanically ventilated sheep (58). In patients undergoing elective cardiothoracic surgery, diaphragm pacing increased diaphragm fiber force with 30% (59). These studies suggest that diaphragm pacing may prevent the development of VIDD.
Other future developments include the modulation of inflammation, proteolytic pathways and oxidative stress to improve diaphragm function. The latter strategies have been explored in experimental animal studies and provide encouraging data (60-63), but first need to be confirmed in humans.

In conclusion, this thesis demonstrates how diaphragm function can be monitored in critically ill patients. In addition, we set first steps towards preventive and therapeutic strategies to optimize diaphragm function. Now is the time to recognize diaphragm dysfunction in critically ill patients and implement monitoring techniques into ICU routines and demonstrate improved outcome with close monitoring of the diaphragm and interventions specifically targeted to enhance diaphragm function.
References


General discussion and future perspectives


Chapter 12

Nederlandse samenvatting, algehele discussie en toekomstperspectieven
Chapter 12

Samenvatting

In de afgelopen decennia is er toenemend bewijs verzameld dat diafragmadisfunctie zich frequent voordoet bij kritisch zieke patiënten dit bijdraagt aan langdurige mechanische beademing. Echter, monitoring van diafragmafunctie tijdens mechanische beademing en tijdens het ontwennen van de beademing heeft weinig aandacht gekregen. Meestal wordt de functie van het diafragma bij kritisch zieke patiënten in zijn geheel niet gemonitord door artsen. Tevens zijn er beperkte interventies beschikbaar die zich specifiek richten op het diafragma. In dit proefschrift zijn ten eerste technieken onderzocht waarmee diafragmafunctie kan worden gemonitord en ten tweede zijn preventieve en therapeutische interventies onderzocht om diafragmafunctie te optimaliseren.

Het eerste deel van dit proefschrift is gericht op het monitoren van diafragmafunctie bij kritisch zieke patiënten. In hoofdstuk 2, een klinisch perspectief, hebben we de voors en tegens besproken van de beschikbare technieken waarmee ademspierfunctie kan worden gemonitord. Deze technieken variëren van gemakkelijk te leren en non-invasieve methoden, zoals het meten van maximale inspiratoire druk en diafragma echografie, tot complexe en geavanceerde metingen, zoals elektrische stimulatie van de nervus phrenicus, elektrische activiteit van het diafragma (EAdi) en transdiafragmale druk (Pdi). Een aantal technieken zijn van beperkte betekenis (zoals fluoroscopie, occlusie druk in de eerste 100 ms) of zijn niet geschikt voor routine monitoring (zoals nervus phrenicus stimulatie). Echter, veelbelovende technieken voor het monitoren van ademspierfunctie zijn registratie van Pdi en EAdi, en diafragma echografie. Deze technieken zijn nu of in de nabije toekomst inzetbaar. Hoewel er geen bewijs is dat monitoring van de ademspieren (en handelen daarop) leidt tot een betere uitkomst voor patiënten, moet naar onze mening het monitoren van de ademspieren evenzeer onderdeel van de klinisch routine zijn als het bewaken van alle andere vitale organfuncties.

Bij kritisch zieke patiënten is het ontwennen van de mechanische beademing een belangrijke fase op intensive care (IC). De overgang van positieve inspiratoire druk door mechanische beademing naar negatieve pleurale druk door spontane ademhaling vraagt veel van de fysiologische reserves van de patiënt. Ontwennen van de beademing faalt wanneer er een disbalans is tussen de ventilatoire behoefte van de patiënt en de capaciteit van het neurale, cardiovasculaire en respiratoire systeem. In hoofdstuk 3 hebben we daarom de pathofysiologie van moeilijke en langdurige ontwenning van de beademing besproken alsmede de technieken die kunnen worden ingezet in het diagnostische traject om de oorzaak van falende ontwenning
Samenvatting

Te achterhalen. Verhoogde luchtwegweerstand en verlaagde compliantie van het respiratoire systeem, cardiale disfunctie, ademspierdisfunctie, cognitieve disfunctie en metabole aandoeningen zijn de belangrijkste oorzaken voor falende ontwenning van de beademing. Het wetenschappelijke onderzoek richt zich voornamelijk op ademspierdisfunctie en cardiale disfunctie als oorzaak voor falende ontwenning van de beademing. Kennis van de complexe pathofysiologie van falende ontwenning in combinatie met een systematische diagnostische aanpak maakt het mogelijk om de primaire oorzaak van falende ontwenning van de beademing te identificeren.

In hoofdstuk 4 is bij een heterogene groep van kritisch zieke patiënten de inspiratoire ademspierfunctie, expiratoire ademspierfunctie en mechanica van de ademhaling tijdens een trial spontaan ademen in detail geanalyseerd. We hebben in dit onderzoek aangetoond dat de expiratoire ademspieren een significante bijdrage (tot 26%) leveren aan de totale arbeid van de ademspieren bij patiënten met een mislukte poging trial spontaan ademen. Daarnaast is de neuromechanische efficiëntie van het diafragma bij patiënten met een mislukte poging trial spontaan ademen 41% lager dan bij patiënten die met succes een trial spontaan ademen ondergingen. Dit bevestigt dat een verminderde capaciteit van het diafragma om inspiratoire druk te genereren, ongeacht de oorzaak hiervan, een belangrijke rol speelt bij falende ontwenning van de mechanische beademing.

In hoofdstuk 5 hebben we bij gezonde proefpersonen aangetoond dat strain en strain rate van het diafragma, berekend middels speckle tracking echografie, sterk gecorreleerd is aan Pdi en EAdi bij een inspiratoire belasting tot 60 cmH\(_2\)O, terwijl verdikking van het diafragma niet gecorreleerd is met Pdi en EAdi. Deze bevindingen suggereren dat geavanceerde technieken voor echografie, zoals speckle tracking, in de toekomst kunnen worden ingezet als non-invasieve methode om diafragmefunctie te monitoren bij kritisch zieke patiënten.

Het tweede deel van dit proefschrift is gericht op preventieve en therapeutische interventies voor het optimaliseren van diafragmefunctie. Het minimaliseren van de schadelijke effecten van mechanische beademing op het diafragma is een belangrijke preventieve maatregel, bijvoorbeeld door het optimaliseren van patiënt-ventilator interactie. Neuromechanically adjusted ventilatory assist (NAVA) is een (non-)invasieve beademingsmodus die de elektrische activiteit van het diafragma gebruikt om de timing en het niveau van drukondersteuning te reguleren.

In hoofdstuk 6 is bij patiënten met COPD middels een automatische analyse van de beademingscurves de patiënt-ventilator interactie gekwantificeerd tijdens verschillende modi van non-invasieve beademing, waaronder NAVA. We hebben aangetoond dat met NAVA beademing de mechanische drukondersteuning in
synchronie is met de neurale inspiratie van de patiënt, terwijl drukgestuurde modi van mechanische beademing geen optimale patiënt-ventilator interactie geven. Tevens stijgt het aantal wasted efforts (gemiste inspiratiepogingen van de beademingsmachine) exponentieel wanneer het verschil in timing tussen de elektrische activiteit van het diafragma en de beademingsmachine meer dan 20% van de inspiratietijd bedraagt. Oftewel, een progressieve mismatch tussen de neurale inspiratie van de patiënt en de mechanische inspiratie van de beademingsmachine is sterk geassocieerd met ernstige vormen van patiënt-ventilator asynchronie. Ondanks deze fysiologische voordelen verbetert NAVA niet de bloedgaswaarden en het comfort van de patiënt.

Bij ARDS patiënten is long-protectieve beademing belangrijk om te voorkómen dat de long nog meer beschadigt. Long-protectieve beademing wordt meestal toegepast door middel van gecontroleerde mechanische beademing. Bij gecontroleerde mechanische beademing is strikte regulatie van het teugvolume en de plateaudruk mogelijk. Deze beademingsmodus vereist echter een hoog sedatie niveau van de patiënt en gaat gepaard met bijwerkingen, in het bijzonder diafragmadisfunctie. Bij ondersteunende modi van mechanische beademing is spontane ademhaling mogelijk, waardoor het risico op diafragmadisfunctie minder is. Echter, ondersteunende mechanische beademing kan weer leiden tot hoge teugvolumes en hoge transpulmonale drukken, waardoor longschade ontstaat. In hoofdstuk 7 is bij patiënten met mild tot matig ARDS het effect onderzocht van verschillende modi van mechanische beademing (drukgecontroleerde beademing [PCV], drukondersteunende beademing [PSV] en NAVA) op parameters van long-protectieve beademing, variabiliteit van de ademhaling en patiënt-ventilator interactie. De hoeveelheid controle die de patiënt kan uitoefenen op de timing en het niveau van ondersteuning van de beademingsmachine is verschillend voor deze drie modi. De belangrijkste conclusie van onze studie was dat het teugvolume en de transpulmonale druk binnen de grenzen van long-protectieve beademing bleven bij een toenemende vrijheid voor de patiënt om de beademingsmachine te controleren. Uitzondering hierop was één patiënt met een ernstige hypercapnische acidose die een teugvolume boven de 10 ml/kg ontwikkelde. Zowel variabiliteit van de ademhaling als patiënt-ventilator interactie verbeterden met NAVA ten opzichte van PCV en PSV. Deze bevindingen suggereren dat er bij patiënten met mild tot matig ARDS het mogelijk is om drukondersteunende beademing toe te passen waarbij het diafragma actief is.

Op basis van de bevindingen in hoofdstuk 7 is een beademingsstrategie ontwikkeld voor ARDS patiënten die hoge teugvolumes ontwikkelen na de overgang van gecontroleerde mechanische beademing naar ondersteunende mechanische
beademing. In een proof-of-concept studie is onderzocht of een lage dosis neuromusculaire blokkers een long-protectieve ondersteunende beademingsstrategie kan faciliteren bij patiënten met longschade en hoge teugvolumes zonder volledig verlies van diafragma activiteit (hoofdstuk 8). Een dergelijke strategie zou mogelijk de voordelen van spontane ademhaling tijdens mechanische beademing kunnen behouden en de risico’s minimaliseren. De resultaten van ons onderzoek tonen aan dat het mogelijke is om partiële spierverslapping van het diafragma zo te titreren dat het teugvolume van 10 ml/kg daalt naar 6 ml/kg met behoud van diafragma activiteit. Gedurende de partiële spierverslapping ontstaan echter wel milde hemodynamisch bijeffecten, waaronder tachycardie en hypertensie, en een hypercapnische acidose. Toekomstige studies moeten zorgvuldig evalueren of partiele spierverslapping van het diafragma mogelijk is gedurende langere tijdsperioden.

In eerder in vitro onderzoek is aangetoond dat levosimendan de calciumgevoeligheid verhoogt van gepermeabilsierde spiervezels van humaan diafragma. Dit heeft tot gevolg dat de spiervezels meer kracht kunnen genereren voor eenzelfde hoeveelheid intracellulair calcium. In hoofdstuk 9 is bij gezonde proefpersonen onderzocht of levosimendan ook in vivo diafragmafunctie verbetert. We hebben aangetoond dat tijdens isometrische belasting van het diafragma, de neuromechanische efficiëntie van het diafragma met ongeveer 20% verbetert. Daarnaast kan levosimendan de afname in kracht van het diafragma door vermoeidheid ongedaan maken.

Systemische inflammatie is een belangrijke risicofactor voor ademspierzwakte. In hoofdstuk 10 is gepoogd om een experimenteel in vivo model te ontwikkelen voor diafragmadisfunctie tijdens systemische inflammatie. In tegenstelling tot diermodellen van systemische inflammatie zagen wij bij gezonde proefpersonen na toediening van een lage dosis endotoxine een toename van de in vivo diafragma contractiliteit in de vroege fase. Gezien deze bevindingen lijkt het humane endotoxemie model niet geschikt voor het testen van interventies om diafragmafunctie verbeteren.
Algehele discussie en toekomstperspectieven

Diafragmadisfunctie komt frequent voor bij kritisch zieke patiënten en is geassocieerd met een verhoogde morbiditeit en mortaliteit. In de studies in dit proefschrift zijn technieken om diafragmamonitoring te monitoren onderzocht alsmede preventieve en therapeutische interventies om diafragmamonitoring te optimaliseren.

Monitoring van diafragmamonitoring tijdens mechanische beademing en het ontwennemen

In de klinische praktijk wordt het diafragma bij kritisch zieke patiënten nauwelijks gemonitord. Beperkte beschikbaarheid en kennis van technieken waarmee je het diafragma kunt monitoren liggen hier aan ten grondslag. In hoofdstuk 2 hebben wij drie technieken geïdentificeerd die veelbelovend zijn om ademspierfunctie te monitoren op de IC: 1) transdiafragmale druk (Pdi), 2) elektrische activiteit van het diafragma (EAdi), en 3) diafragma echografie. Deze technieken zijn in de afgelopen jaren in toenemende mate onderzocht.

Door het meten van Pdi kun je de drukopbouw van het diafragma heel specifiek monitoren. Deze techniek vereist het plaatsen van een dubbele ballon katheter (maag en oesophagus ballon). De daling van druk in de oesophagus (Pes) tijdens inspiratie representeren de resulterende actie van alle ademspieren en vereist slechts het plaatsen van één ballon. Deze benadering is voor het monitoren van ademspieractiviteit bij kritisch zieke patiënten uitvoerig beschreven (1-3). Bij beademde patiënten met spontane ademhaling wordt Pes monitoring ook gebruikt om de arbeid van de ademspieren en patiënt-ventilator interactie te monitoren.

In hoofdstuk 7 en hoofdstuk 8 zien we dat de daling in Pes tijdens inspiratie kan variëren van laag tot zeer hoog. Dit betekent dat de activiteit van de ademspieren varieert van onvoldoende tot bovenmatig. Zowel onvoldoende als bovenmatige inspiratoire spieractiviteit kan leiden tot diafragmadisfunctie (4-9). Monitoring van Pes of Pdi tijdens mechanische beademing maakt het mogelijk om de ondersteuning van de beademingsmachine te titreren tot een gewenste activiteit van de ademspieren. Een door de ademspieren gegenereerde druk tussen de 5 en 10 cmH2O valt binnen het fysiologisch bereik (10). Door het monitoren van Pes tijdens een trial spontaan ademen kan onderscheid worden gemaakt tussen succesvolle en falende ontwennening (11). In hoofdstuk 4 zijn bij patiënten tijdens een trial spontaan ademen de veranderingen in Pes en maagdruk (Pga) gemeten. Daarnaast hebben we Pes en Pga gebruikt om meer geavanceerde parameters te berekenen, zoals intrinsieke positief eind-expiratoire druk, expiratoire stijging van de maag druk, ademarbeid en...
Algemeen discussie en toekomstperspectieven

pressure-time product. Vanuit een diagnostisch perspectief geven deze parameters relevante informatie om de oorzaak van falende ontwenning voor een individuele patiënt te bepalen (12, 13). Voor routine monitoring van de ademspieren vereist deze gedetailleerde analyse echter een te hoog niveau van deskundigheid. Monitoring van enkel de drukverandering van Pes of Pdi tijdens inspiratie is beter geschikt voor een routinematige klinische toepassing. Verschillende studies hebben aangetoond dat Pes monitoring kan worden gebruikt om patiënt-ventilator interactie te monitoren (14-16). In dit proefschrift hebben patiënt-ventilator interactie echter gemonitord met behulp van de EAdi, zoals beschreven in de volgende paragraaf.

Een andere toepassing van Pes monitoring, naast het monitoren van de ademspieren, is het sturen van long-protectieve mechanische beademing bij patiënten met acute respiratory distress syndrome (ARDS). Wij hebben laten zien dat Pes (in de berekening van de transpulmonale druk) kan worden gebruikt om veilige of schadelijk drukken voor de long te identificeren tijdens mechanische beademing bij ARDS patiënten (hoofdstuk 7-8).

Monitoring van de elektrische activiteit van het diafragma (EAdi) is de standaard om patiënt-ventilator interactie te evalueren (17-22). Vaak wordt patiënt-ventilator interactie handmatig gescoord. Daarentegen hebben wij patiënt-ventilator interactie gèrevalueerd met behulp van een geautomatiseerd algoritme (op basis van de EAdi) bij patiënten met chronic obstructive pulmonary disease (COPD) tijdens non-invasieve beademing (hoofdstuk 6) en bij ARDS patiënten tijdens invasive beademing (hoofdstuk 7). Deze geautomatiseerde analyse detecteert asynchronieën, zoals wasted efforts, maar maakt het ook makkelijker om meer subtile dissynchronieën (timing fouten), zoals trigger vertragingen en cycling-off fouten te detecteren. Dissynchronieën worden gemakkelijk gemist bij een visuele inspectie van de beademingscurven, maar kunnen zeker klinisch relevant zijn. Het aantal wasted efforts stijgt namelijk sterk wanneer dissynchronieën oplopen tot meer dan 20% van de respiratoire cyclus (hoofdstuk 6). Een geautomatiseerde analyse van EAdi (of Pes) zou in beademingsmachines kunnen worden ingebouwd om monitoring van patiënt-ventilator interactie te vereenvoudigen.

Het EAdi signaal kan ook worden gebruikt om de neurale aansturing van de ademspieren te monitoren tijdens mechanische beademing en het ontwennen. Wij hebben aangetoond bij ARDS patiënten die werden beademd met 6 ml/kg dat er een grote variatie kan zijn in de mate van neurale aansturing van de ademspieren (hoofdstuk 7). Dit suggereert dat het instellen van lage teugvolumes op de beademingsmachine kan leiden een groot verschil in adempijkkel tussen patiënten. Dit is waarschijnlijk het gevolg van de complexe interactie tussen sedatie,
beademingsmodus en het niveau van ondersteuning van de beademingsmachine. Het monitoren van EAdi kan in deze situaties helpen om beademing van ARDS patiënten te optimaliseren en daarmee onvoldoende of bovenmatige ondersteuning van de beademingsmachine te voorkómen. Een nadeel van het monitoren van EAdi is dat de amplitude van het signaal sterk wordt beïnvloed door de individuele anatomie van de patiënt met als gevolg dat het vergelijken tussen patiënten moeilijk is. Bovendien is er geen methode om de optimale EAdi amplitude te bepalen voor een individuele patiënt. Recentelijk zijn er pogingen gedaan om ademarbeid te schatten met behulp van het EAdi signaal (23) en om de bijdrage van de patiënt aan het teugvolume tijdens mechanische beademing te berekenen (24). Echter, de betrouwbaarheid van deze metingen is slecht (25) en daarom nog niet klinische toepasbaar. Nieuw onderzoek is vereist om een betrouwbare methode te ontwikkelen die kan vaststellen wat de optimale EAdi amplitude is voor een individuele patiënt.

De verhouding tussen Pdi en EAdi representeert de neuromechanische efficiëntie (NME) van het diafragma, oftewel het vermogen van het diafragma om neurale respiratoire input om te zetten naar mechanische respiratoire output. Een geleidelijke afname in NME over een aantal dagen kan betekenen dat het diafragma verzwakt of vermoeid raakt, terwijl een toename herstel suggereert. In ons onderzoek naar ontwenning van de beademing is aangetoond dat de NME lager is bij falende ontwenning dan bij succesvolle ontwenning (hoofdstuk 4). In deze studie is de NME berekend met behulp van een dubbele ballon katheter met meerdere elektroden zodat continu Pdi en EAdi kon worden berekend. In eerder onderzoek is NME van de ademspieren berekent als de verhouding tussen de daling in monddruk tijdens een inspiratoire occlusie en EAdi (26-28). Met deze methode is aangetoond dat NME falende ontwenning van de beademing kan voorspellen (26). In een kleine gerandomiseerde klinische studie werd aangetoond dat beademing met NAVA zorgt voor een verbetering in NME terwijl dit niet het geval is bij beademing met PSV (27). Dit suggereert dat het monitoren van NME zinvol kan zijn om veranderingen in efficiëntie van de ademspieren te detecteren tijdens mechanische beademing. Echter, ander onderzoek toont aan dat de (trend van) NME niet geassocieerd is met risicofactoren voor diafragmadisfunctie of klinische uitkomst (28). Nader onderzoek is nodig naar de waarde van het monitoren van NME tijdens mechanische beademing en ontwenning en de meest optimale en praktische werkwijze.

Naast fysiologische metingen neemt de populariteit van beeldvorming van het diafragma met echografie toe om diafragmadisfunctie te monitoren op de IC. Met echografie kan de dikte, verdikking tijdens inspiratie en de verplaatsing van het diafragma worden bepaald (29). Eerder onderzoek heeft aangetoond dat bij
kritisch zieke patiënten de dikte van het diafragma afneemt tijdens een periode van mechanische beademing en dat de verdikking van het diafragma afhankelijk is van de ondersteuning die de beademingsmachine geeft (9, 30). Echter, correlaties tussen functionele metingen (zoals Pdi) en verdikking van het diafragma zijn laag (31). Daarom is er behoefte aan nieuwe echografie technieken. Speckle tracking imaging van het diafragma, een geavanceerde echografie techniek, laat wel een correlatie zien met Pdi (hoofdstuk 5) en kan mogelijk in de toekomst worden ingezet als non-invasieve methode om ademspierfunctie te monitoren bij kritisch zieke patiënten.

Dit proefschrift laat zien dat monitoring van het diafragma toepasbaar is bij kritisch zieke patiënten. Monitoring van Pdi (of Pes), EAdi en diafragma echografie geven complementerende informatie over het diafragma. Idealiter beschikken artsen over alle drie de technieken, zoals bloeddruk, elektrocardiografie en echocardiografie gebruikt worden voor monitoring van cardiale functie. Echter het gebruik van één van deze technieken bij de dagelijkse zorg voor IC patiënten zou al een grote stap voorwaarts zijn voor het diafragma. Het monitoren van ademspierfunctie op de IC zou evenzeer onderdeel van de routine moeten zijn als het monitoren van enig andere orgaan functie. Zorgvuldige monitoring van diafragmafunktie is een voorwaarde om veranderingen te kunnen detecteren, maar monitoring is zinloos wanneer er geen actie kan worden ondernomen om diafragmadisfunctie te voorkómen of om diafragmafunktie te verbeteren. Om deze reden zijn in dit proefschrift ook mogelijke interventies onderzocht.

**Preventieve en therapeutische interventies om diafragmafunktie te optimaliseren**

Er zijn momenteel beperkt interventies beschikbaar om diafragmafunktie te optimaliseren bij kritisch zieke patiënten (32). In dit proefschrift zijn nieuwe interventies gericht op het voorkómen van diafragmadisfunctie (hoofdstuk 6-8) en het verbeteren van diafragmafunktie (hoofdstuk 9-10) geëvalueerd.

Mechanisch beademing is een belangrijke factor die bijdraagt aan diafragmadisfunctie bij kritisch zieke patiënten, dit wordt ook wel beademings-geïnduceerde diafragmadisfunctie (VIDD) genoemd (33, 34). Inactiviteit van het diafragma tijdens mechanische beademing is de belangrijkste pathofysiologische trigger die leidt tot VIDD, vooral tijdens gecontroleerde mechanische beademing (34). Hoge niveaus van ondersteuning tijdens ondersteunende beademing kunnen echter ook diafragmadisfunctie veroorzaken (8). Om diafragma inactiviteit te beperken tijdens mechanische beademing kunnen preventieve maatregelen worden getroffen.
Neurally adjusted ventilatory assist (NAVA) is een beademingsmodus die de elektrische activiteit van het diafragma gebruikt om de beademingsmachine aan te sturen (35). Bij COPD patiënten en ARDS patiënten verbetert NAVA de patiënt-ventilator interactie in vergelijking met drukgestuurde beademing (PSV) (hoofdstuk 6-7). Een slechte patiënt-ventilator interactie is sterk geassocieerd met langdurige mechanische beademing (36, 37). NAVA kan daardoor potentieel de duur van mechanische beademing verkorten en daarmee de uitkomst van beademende patiënten verbeteren.

De proportionaliteit tussen EAdi en het niveau van ondersteuning bij NAVA voorkomt tevens bovenmatige ondersteuning, zoals het geval kan zijn bij PSV (38-41). Ondanks deze fysiologische voordelen van NAVA, zien wij in onze onderzoeken geen verbeteringen in bloedgaswaarden en patiënt comfort met NAVA (hoofdstuk 6-7). Dit laatste kan mogelijk het gevolg zijn van de korte periode (30 min) waarin wij de verschillende beademingsmodi hebben onderzocht. Onlangs werd in een multicenter gerandomiseerde gecontroleerde trial aangetoond dat NAVA een veilige en toepasbare beademingsmodus is over een langere periode, maar dat het niet de ligduur op de IC of sterfte vermindert (42). Deze trial is uitgevoerd in een heterogene groep van kritisch zieke patiënten met acuut respiratoir falen, terwijl voornamelijk bepaalde groepen, zoals COPD patiënten of patiënten met hoge niveaus van ondersteuning, fysiologisch baat lijken te hebben bij NAVA. Nieuwe studies zijn nodig om te bepalen of en bij welke patiëntengroepen NAVA diafragmadisfunctie kan voorkómen en de uitkomst van patiënten kan verbeteren.

Om activiteit van het diafragma te stimuleren, moeten beademde patiënten zo spoedig mogelijk worden beademd met een ondersteunende beademingsmodus. Bij patiënten met ARDS kan ondersteunende beademing echter het risico verhogen op hoge teugvolumes en hoge transpulmonale drukken (P_L) en daarmee longschade verergeren, dit wordt ook wel beademings-geïnduceerde longschade genoemd (43). Het gebruik van ondersteunende beademing en het moment waarop dit moet worden gestart bij ARDS patiënten staat ter discussie (44-47). In hoofdstuk 7 is aangetoond dat bij geselecteerde patiënten met een mild tot matig ARDS ondersteunende mechanische beademing (NAVA en PSV) mogelijk is binnen de grenzen van long-protectieve beademing. Echter, er zijn ook patiënten in deze studie met een te hoog teugvolume en P_L. In deze specifieke casus, waar ondersteunende mechanische beademing faalt, zullen artsen zeer waarschijnlijk overgaan op gecontroleerde beademing en/of een hoog niveau van sedatie nastreven. Als alternatief kan partiele spierverslapping van het diafragma met behulp van een lage dosis rocuronium ook een laag teugvolume en P_L faciliteren tijdens ondersteunde mechanische beademing en met behoud van diafragma activiteit (hoofdstuk 8). Deze nieuwe beademingsstrategie
bij ARDS patiënten combineert daardoor long-protectieve beademing en diafragma-protectieve beademing. Een nadeel van deze beademingsstrategie is het optreden van milde hemodynamisch bijeffecten, waaronder tachycardie en hypertensie, en een hypercapnische acidose. Om hemodynamische veranderingen te minimaliseren, zou de snelheid van toename in PaCO₂ kunnen worden gemanipuleerd door rocuronium meer geleidelijk te titreren. Nader onderzoek moet zich richten op het begrijpen van de gevolgen van partiële spierverslapping voor diafragmafunctie op de lange termijn en de haalbaarheid van deze strategie onder lage niveaus van sedatie.

Momenteel zijn er geen farmacologische middelen beschikbaar om de functie van het diafragma te verbeteren bij kritisch zieke patiënten. Levosimendan is een middel dat de gevoeligheid voor calcium in de spiercel verhoogt en staat geregistreerd als middel om de contractiliteit van het hart te verhogen bij patiënten met acuut hartfalen (48). In eerder in vitro onderzoek is aangetoond dat levosimendan ook de calciumgevoeligheid van gepermeabiliseerde spiervezels van humaan diafragma verhoogt (49). Daarnaast reduceert levosimendan eiwit nitrosylering en markers van oxidatieve stress bij knaagdieren in een model van systemische inflammatie (50). In dit proefschrift hebben we aangetoond dat toediening van levosimendan aan gezonde proefpersonen de contractiliteit en neuromechanische efficiëntie van het diafragma verbetert tijdens ademen tegen een hoge belasting (hoofdstuk 9). Onze bevindingen suggereren een mogelijkheid voor nieuwe therapeutische behandeling van diafragmadisfunctie bij kritisch zieke patiënten. Ook bij onderzoek naar een alternatieve troponine activator is gebleken dat de calciumgevoeligheid van gepermeabiliseerde spiervezels van humaan diafragma kan worden verhoogd (51), maar dit is nog niet bevestigd in humane studies. Recentelijk is er in een grote gerandomiseerde gecontroleerde trial onderzocht of levosimendan de Ernst van orgaanfalen bij volwassenen met sepsis vermindert (52). In schijnbare tegenstelling tot onze bevindingen, blijkt dat levosimendan is geassocieerd met een lagere kans op een succesvolle ontwenning van de beademing (52). Dit onderzoek was echter niet specifiek gericht op het evalueren van het effect van levosimendan op ontwenning van de beademing of het verbeteren van diafragmafunctie. In deze studie werd levosimendan in de acute fase van een septische shock gegeven om de inotrope en anti-inflammatoire effecten te onderzoeken. Om de effecten op diafragmafunctie te evalueren kan levosimendan beter worden toegediend aan patiënten met een moeilijke ontwenning van de beademing, na de acute fase. Op dit moment onderzoeken we de effecten van levosimendan op diafragmafunctie in een gerandomiseerde gecontroleerde trial bij kritisch zieke patiënten (ClinicalTrials.gov NCT01721434).
In aanvulling op het werk in dit proefschrift zijn er in de afgelopen jaren andere interventies onderzocht gericht op het diafragma. In het bijzonder zijn er studies verricht naar de effecten van inspiratoire spierkracht training (IMST) en pacing van het diafragma. Drie gerandomiseerde studies hebben de effecten van IMST bij beademde kritisch zieke patiënten geëvalueerd (53-55). Deze studies hebben met verschillende trainingsprotocollen gevonden dat IMST de maximale inspiratoire druk verbetert en dat er geen bijwerkingen van IMST zijn. Bovendien liet twee studies zien dat de tijd tot ontwenning van de beademing werd verkort (53, 54). Dus IMST wordt getolereerd door kritisch zieke patiënten met ademspierzwakte en het lijkt de duur van de beademing te kunnen verkorten. Nieuwe studies zijn nodig om te bepalen wat het optimale trainingsprotocol is en wanneer en bij wie gestart kan worden met IMST.

Pacing van het diafragma gebeurt door middel van elektrische stimulatie van de nervus phrenicus via een intrathoracale of intradiafragmale benadering. Diafragma pacing via implantatie van elektroden is momenteel alleen geïndiceerd bij tetraplegische patiënten met een hoge dwarslaesie of patiënten met permanente of slaap-gerelateerde centrale hypoventilatie (56). Het doel van diafragma pacing bij deze patiënten is het verminderen van de afhankelijkheid van (voornamelijk nachtelijke) mechanische beademing. Bij kritisch zieke beademde patiënten kan tijdelijke diafragma pacing via een transveneuze benadering mogelijk diafragmadisfunctie voorkómen of zelfs de contractiliteit van het diafragma verbeteren. Recent is in een proefdierstudie aangetoond dat bij vroege transveneuze diafragma pacing de dikte van het diafragma en de dwarsdoorsnede van spiervezels behouden blijft bij beademde varkens met diafragma pacing versus beademde varkens zonder diafragma pacing (57). Preventie van diafragma atrofie door middel van diafragma pacing is ook aangetoond in een preliminaire studie bij drie beademde schapen (58). Bij patiënten die electieve cardiothoracale chirurgie ondergingen verhoogde diafragma pacing de kracht van diafragma vezels met 30% (59). Deze studies suggereren dat diafragma pacing de ontwikkeling van VIDD in de toekomst kan voorkómen.

Andere ontwikkelingen om diafragmafunctie te verbeteren zijn de modulatie van inflammatie, proteolytische pathways en oxidatieve stress. In exploratief onderzoek bij dieren worden positieve resultaten geboekt (60-63), maar deze moeten nog worden bevestigd in humaan onderzoek.
Tot slot, dit proefschrift toont aan hoe diafragmefunctie kan worden gemonitord bij kritisch zieke patiënten. Daarnaast hebben we eerste stappen gezet richting preventieve en therapeutische strategieën om diafragmefunctie te optimaliseren. De tijd is daar om diafragmadisfunctie bij kritisch zieke patiënten te erkennen en monitoring technieken te implementeren in de routines van de IC en te demonstreren dat zorgvuldige monitoring van het diafragma en gerichte interventies om diafragmefunctie te verbeteren leiden tot een betere uitkomst voor patiënten.
Referenties


Algehele discussie en toekomstperspectieven
Dankwoord

Met mijn promotie komt er een einde aan een mooie en leerzame periode van mijn carrière. Velen hebben bijgedragen aan de totstandkoming van dit proefschrift.

Allereerst wil ik alle gezonde vrijwilligers, patiënten en hun families die toestemming gaven om deel te nemen aan mijn onderzoek bedanken.

Prof. dr. Heunks, beste Leo, ik heb het ontzettend getroffen met jou als promotor. Je hebt me in alle facetten van de wetenschap wegwijs gemaakt. Dank voor het vertrouwen, de waardevolle correcties van mijn manuscripten, de diepgaande discussies, het perfectioneren van presentaties en de ruimte om mezelf te kunnen ontwikkelen. Jouw enthousiasme en gedrevenheid hebben mij altijd weten te motiveren op lastige momenten. Naast alle serieuze kwesties heb je ook veel plezier in mijn promotie gebracht. Je voorliefde voor voetbal en de rivaliteit tussen mijn Ajax en die andere club van jou, leverde elke maandagochtend weer mooie gesprekken op. Ondanks onze eigen transfers, hoop ik dat we in de toekomst blijven samenwerken.

Prof. dr. Van der Hoeven, beste Hans, als promotor en afdelingshoofd zijn jouw inspirerende, bemoedigende en wijze woorden altijd van grote waarde geweest. Ik heb veel geleerd van de discussies die we hadden over mijn manuscripten en de correcties die je aandroeg. Ik wil je bedanken voor het vertrouwen dat je vanaf het eerste moment hebt gehad in mijn opleiding en beroep. Je hebt je altijd hard gemaakt voor de klinische rol van de technisch geneeskundige. Ik heb met veel trots op de IC gewerkt.

Dr. Van Hees, beste Jeroen, je hebt als copromotor een belangrijke rol gespeeld in de totstandkoming van dit proefschrift. Jouw kritische kijk op de studies, resultaten en manuscripten hebben me altijd weer tot nadenken gezet en daar heb ik veel van geleerd. Je kennis van de spierfysiologie in combinatie met je gevoel voor humor zorgden altijd voor interessante en leuke discussies. Ons gezamenlijk hoogtepunt was natuurlijk het geven van een sunrise seminar in Philadelphia, niemand kan ontkennen dat het een groot succes was.

Dr. Sinderby and dr. Beck, dear Christer and Jennifer, after having read all your great articles on diaphragm electromyography I was honoured you got involved in my research. You have learned me a great deal on the physiology of breathing and your revisions significantly improved my manuscripts. I have had a wonderful time in
Toronto discussing our data day and night. Dear Norman, your technical assistance with the measurement setup and data analysis were of indispensable value. Besides talking business, I really enjoyed riding our bikes together on the endless Canadian roads. Christer, Jennifer and Norm, you are truly an amazing team. Thank you for all your support and hospitality.

Prof. dr. ir. Stegeman, beste Dick, jouw expertise op het gebied van de elektromyografie is zeer waardevol geweest bij de start van mijn promotie. Ik vind het een eer dat ik jou heb mogen opvolgen bij de Klinische Neurofysiologie. Bedankt voor het vertrouwen.


Al mijn collega onderzoekers op de IC wil ik vooral bedanken voor het werkplezier dat ik elke dag heb gehad. In de afgelopen jaren zijn er veel promovendi gekomen en gegaan, maar de sfeer op de afdeling was altijd geweldig. Een ideale balans tussen inspanning en ontspanning. In het bijzonder wil ik een aantal collega’s bedanken. Matthijs en Mark, mijn kamergenoten voor het grootste deel van mijn promotie, bedankt voor het vele lachen, de mooie gesprekken en de fietstochtjes. Matthijs, je loopt altijd een paar jaar voor op mij in je carrière en privé. Bedankt voor al je adviezen daarin van klein tot groot. Mark, bedankt voor je aanstekelijke enthousiasme, hulp bij statistische vragen en culinaire tips. Lucas, je bent een ontzettend fijne en betrokken collega. Ik heb veel geleerd van de manier waarop jij studenten begeleid. Onze survivaltochten in de Duitse bossen waren fantastisch. Willem-Jan, als collega diafragma onderzoeker heb ik van jouw studies veel geleerd over de biochemische en moleculaire spierfysiologie. De congressen in de VS waar we vaak de kamer deelden waren altijd erg gezellig. Lisanne, bedankt voor de talloze metingen die je voor me hebt uitgevoerd tot ‘s avonds laat. Het is mooi om te zien dat je onmisbaar bent geworden op de IC.

Naast mijn fijne collega’s wil ik mijn vrienden en familie bedanken voor de noodzakelijke ontspanning, relativering en steun.
Kevin, de verschillen tussen jouw snelle zaken en mijn trage wetenschap zijn groot, maar onder het genot van een biertje vinden we verassend vaak parallellen. Het aantal fietstochten en avonden in de kroeg zijn minder dan in Enschede, maar ik geniet er nog net zoveel van. Thomas en Jasper, na al die jaren hebben we nog net zo veel lol als vroeger. Soms met zijn drieën, maar inmiddels ook met zijn elven. Hoe mooi is dat. De tijd vliegt, over drie jaar is het al 2020.


Henk en Christl, jullie zijn staan altijd voor Janet en mij klaar. Bedankt voor de vele momenten dat jullie op Luuk hebben gepast, zodat papa nog even kon gaan werken aan zijn proefschrift. Anita en Jeff, bedankt voor jullie interesse in mijn onderzoek en de leuke momenten samen met Véronique en Elena.

Janine, grote zus, jij bent mij voorgegaan in de wetenschap en met promoveren. Het is ontzettend fijn om een zus te hebben waarmee je de vreugde en frustraties van het onderzoek kunt delen. Laten we snel samen een artikel schrijven. Remy, bedankt voor je oprechte interesse. Lieve Jaslynn, mocht je geïnteresseerd zijn, ik leg het je ooit graag allemaal uit.


Lieve Luuk, jij gaat ook elke dag op onderzoek uit en daar geniet ik volop van. Lieve Janet, de laatste woorden van dit dankwoord zijn voor jou. Meer dan je beseft ben je belangrijk geweest voor mij, als steun bij tegenslagen en bij het vieren van successen. Ik ben ontzettend gelukkig met jou en Luuk.
List of publications


Curriculum vitae


Naast zijn professionele werkzaamheden is Jonne medeoprichter van de beroepsvereniging voor technische geneeskundigen, de Nederlandse Vereniging voor Technische Geneeskunde, en heeft van 2009 tot 2013 de taak van voorzitter vervuld.

Jonne is getrouwd met Janet Doorduin - van Delden. Samen hebben zij een zoon, Luuk.