Societal impact of sepsis and inflammation in the ICU and beyond

Maria Elizabet Koster-Brouwer

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Societal impact of sepsis and inflammation in the ICU and beyond

Maatschappelijke impact van sepsis en inflammatie tijdens en na IC-verblijf

(met een samenvatting in het Nederlands)

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Maria Elizabet Brouwer
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te Urk

Promotor: Prof. dr. M.J.M. Bonten

copromotor: Dr. O.L. Cremer

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Chapter 1

General introduction

Infections, induced by an invading pathogen, are one of the most common causes of morbidity and mortality worldwide (1). The human response towards such invading pathogens may lead to sepsis. In patients with sepsis a misbalance in pro- and anti-inflammatory markers (e.g., tumor necrosis factor-α, interleukin-6, and interleukin-10) is observed (2, 3). This unbalanced inflammatory response might affect the functioning of organ systems, for example leading to acute respiratory failure, renal dysfunction, or shock (2, 4-6). According to the most recent consensus definition sepsis is "a life-threatening organ dysfunction caused by a dysregulated host-response to infection" (5).

Hence, patients with sepsis need immediate treatment to combat the infection and to reverse life-threatening organ dysfunction (7). However, accurate and timely diagnosis of sepsis is challenged by the fact that sepsis is a so-called syndromal diagnosis. There is a large variety in signs and symptoms of sepsis depending on affected organ systems, body site of infection, and timing of presentation to the ICU. This might often result in misdiagnosis. In more than 40% of ICU patients admitted with a clinical suspicion of sepsis and treated with antimicrobial therapy, infection could later be ruled-out or was only 'possible' on post-hoc assessment using all available data of the ICU admission (8). Furthermore, when using all available clinical data and standardized definitions to diagnose infection, overall interobserver agreement was high (89%), but variability between observers was seen in determining source, plausibility and causative pathogen of infection (9).

Considering this diagnostic difficulty, numerous approaches have been investigated to expedite the diagnostic process, for example the use of single biomarkers or transcriptomics (10, 11). However, using a single biomarker might be a too simplified representation of the complex host-immune response during sepsis (12, 13). Therefore, research has focussed on the use of novel techniques, for example transcriptomics; the analysis of host gene expression using RNA transcripts (11). In Part I of this thesis, we explored whether such a novel test could lead to improved diagnosis in critically ill patients with sepsis.

Sepsis as a global threat

In 2017, the World Health Organization presented a resolution in which sepsis is stated as a threat for global health. This resolution emphasizes the importance of improved prevention, diagnosis and management of sepsis (14). In a recent systematic review assessing the impact of sepsis, the incidence of treated sepsis cases was reported to be 288 per 100,000 person years annually. The overall case fatality rate was 21% (range 5-42.5%). These numbers imply that more than 5 million persons die due to sepsis globally each year (15). In the ICU, sepsis is observed in almost 30% of all patients; in 18% sepsis is already present at admission, and 12% develops sepsis during admission (16). In the Netherlands, 9000 patients are admitted to an ICU with sepsis, representing

11% of all ICU admissions (17), and 3500 patients die of sepsis each year, making it a common cause of death (18).

Risk factors to develop sepsis are higher age, comorbidities, impaired immune status, and acute events such as burns or trauma (4, 19, 20). Therefore, the trends of aging and multimorbidity in the global population are likely to lead to an increase of ICU admissions and sepsis episodes, and to pressure health care budgets in the nearby future (21, 22). Therefore, in Part II of this thesis we estimated the economic burden associated with sepsis in the Netherlands.

Post-intensive care syndrome

Although sepsis is still one of the leading causes of death worldwide, the case fatality rate has decreased over the last years. Hence, the proportion of ICU survivors in the general population has increased (23-25), and the focus of research has shifted from short-term mortality to long-term mortality and morbidity. By virtue of this type of research, it is now known that ICU survivors are faced with diminished health after ICU discharge due to physical and psychological disability. For example, ICU survivors are confronted with generalized weakness, cardiovascular problems, decreased mobility, anxiety and depression, and cognitive problems (26, 27). These problems can be present up to years after ICU discharge. The long-term consequences of critical illness on the cognitive, psychological and physical domain have been coined as the post-intensive care syndrome (PICS) (26).

Risk factors associated with poor long-term health outcomes in ICU survivors have similarities with risk factors for sepsis, for example frailty prior to ICU admission, higher age, and comorbidities (24, 26). This is reflected in a high incidence of PICS in this population: after surviving a sepsis episode in hospital, approximately 33% of patients die during the first year after discharge (half of these deaths are estimated to be sepsis-related), 50% reach their previous level of functioning, and the remaining 17% of patients suffers from PICS (25, 28).

Patients surviving sepsis have a risk of approximately 25% to be re-hospitalized in the first month after hospital discharge (24, 29). Remarkably, unresolved or recurrent infections are the most common reason for these new hospital episodes, suggesting that there might be residual immunosuppression (or at least inflammatory changes) making them more vulnerable to infection (3, 24, 25, 30-32). There might also be a higher susceptibility to diseases related to inflammatory processes, such as metabolic disorders, cardiovascular diseases, and other age-related diseases (33-35). All together, these long-term health care problems might lead to reduced health-related quality of life and high health care associated resource use in this population of sepsis survivors (36, 37). Therefore, Part III of the research in this thesis was focussed on exploring the etiologic and prognostic association between sepsis, inflammation and chronic health care status.

Thesis objective

The general aim of this thesis was to estimate to what extent sepsis and inflammation impact society, both during and following an ICU admission. Specific objectives were to (1) explore the clinical value of a novel diagnostic test for sepsis, (2) estimate the economic burden of sepsis in the Netherlands, and (3) model long-term sequelae of sepsis and inflammation in the ICU.

Setting

Some studies in this thesis were embedded in the Molecular Diagnosis and Risk Stratification of Sepsis (MARS) project. Starting in 2010, the aim of this project was to develop and improve tools supporting accurate diagnosis and prognosis of patients at risk for sepsis in the ICU. To this end, demographic and clinical data were prospectively collected by trained researchers alongside storage of left-over blood plasma in a biobank in the ICUs of the Academic Medical Center Amsterdam and the University Medical Center in Utrecht (UMCU). For one study, part of these data was merged with data from the Achmea Health Database, a database containing information on reimbursed health care of more than 4 million Dutch citizens over prolonged periods of time. Data on long-term outcomes used in this thesis were routinely collected using a survey. This follow-up questionnaire was sent to ICU survivors of the UMCU approximately one year after ICU discharge. The survey covers several topics regarding long-term health, including questions on current medication use, physical functioning, health-related quality of life, anxiety, depression, and pain.

Outline

In the first part of this thesis we aimed to investigate to what extent a new test to diagnose infection in the ICU could be of clinical benefit by expediting sepsis diagnosis. In **Chapter 2** we report a first clinical validation study evaluating the diagnostic performance of the SeptiCyte LAB test. Author correspondence regarding this study is included in **the addendum to Chapter 2.**

In the second part of this thesis we aimed to report sepsis-associated health care costs in the ICU and after ICU discharge. In **Chapter 3** we estimated direct medical costs – including costs for diagnostic procedures, therapeutic interventions, and accommodation – using a bottom-up micro-costing approach in patients admitted to the ICU with sepsis or septic shock. We also investigated whether these acute health care costs could be predicted using patient information available at the time of ICU admission. For **Chapter 4** we combined data from the MARS-study with data from the Achmea Health Database. We investigated to what extent health care utilization and associated costs changed after an ICU admission for sepsis when comparing it to the 2-year period before hospitalization. We also assessed the correlation of health care costs with health-related quality of life.

The third part of this thesis consists of three studies regarding modelling of long-term sequelae of sepsis and inflammation in the ICU. Using a matched case-control design, we explored the etiologic association between severe inflammation and tissue hypoxia in the ICU and the development of type 2 diabetes mellitus in the year after ICU discharge in **Chapter 5**. A prognostic approach was chosen in **Chapter 6**, a study in which we described the incidence and characteristics of newly-acquired chronic pain in ICU survivors and tried to predict which patient will develop chronic pain using information available at ICU discharge. In **Chapter 7** a methodological study is presented on the topic of proportional hazards assumption in survival analysis. In this chapter we aimed to demonstrate by which mechanisms this assumption can be violated in clinical studies and that it might influence conclusions about the studied effect.

Finally, in **Chapter 8** we summarize the main findings of this thesis and address the implications of our research for society and the future research agenda.

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Lieve tante Marjan,
Vanharte beterschap
hoop dat u snel
weer opknapt
liefs Johan. Heleen
en de kids

Part I

Diagnosing infection

2

Chapter 2

Validation of a novel molecular host response assay to diagnose infection in hospitalized patients admitted to the ICU with acute respiratory failure

Maria E. Koster-Brouwer
Diana M. Verboom
Brendon P. Scicluna
Kirsten van de Groep
Jos F. Frencken
Davy Janssen
Rob Schuurman
Marcus J. Schultz
Tom van der Poll
Marc J.M. Bonten
Olaf L. Cremer
On behalf of the MARS consortium

Published in Critical Care Medicine (2018)

Abstract

Objective Discrimination between infectious and non-infectious causes of acute respiratory failure (ARF) is difficult in patients admitted to the intensive care unit (ICU) after a period of hospitalization. Using a novel biomarker test (SeptiCyte LAB) we aimed to distinguish between infection and inflammation in this population.

Design Nested cohort study.

Setting Two tertiary mixed ICUs in the Netherlands.

Patients Hospitalized patients with ARF requiring mechanical ventilation upon ICU admission from 2011 to 2013. Patients having an established infection diagnosis or an evidently non-infectious reason for intubation were excluded.

Interventions None.

Measurements and Main Results Blood samples were collected upon ICU admission. Test results were categorized into four probability bands (higher bands indicating higher infection probability) and compared with the infection plausibility as rated by post-hoc assessment using strict definitions. Of 467 included patients, 373 (80%) were treated for a suspected infection at admission. Infection plausibility was classified as ruled-out, undetermined, or confirmed in 135 (29%), 135 (29%), and 197 (42%) patients, respectively. Test results correlated with infection plausibility (Spearman's rho 0.332; $p < 0.001$). After exclusion of undetermined cases, positive predictive values were 29%, 54%, and 76% for probability bands 2, 3, and 4, respectively, whereas the negative predictive value for band 1 was 76%. Diagnostic discrimination of SeptiCyte LAB and CRP was similar ($p=0.919$).

Conclusion Among hospitalized patients admitted to the ICU with clinical uncertainty regarding the etiology of ARF, the diagnostic value of SeptiCyte LAB was limited.

Introduction

Numerous biomarkers have been evaluated for diagnostic utility in distinguishing infection from sterile inflammation in critically ill patients, including C-reactive protein (CRP), procalcitonin, and several coagulation markers (1, 2). Despite the clear association of these biomarkers with the presence of systemic inflammation, most did not diagnose or rule-out infection with sufficient rigor (1-4). Distinct protein biomarkers likely provide an (over)simplified representation of the host immune response to infection (2, 5), which is very complex yet largely similar to that following major surgery, trauma, and various other diseases triggering systemic inflammation (6). As a result, the use of single biomarkers may be predestined to yield only limited diagnostic value (2, 5).

Recently, a novel diagnostic test (SeptiCyte LAB, Immunexpress, Seatle, WA) was developed which aims to provide a probability of infection based on the expression of a specific genomic fingerprint consisting of CEACAM4 (carcinoembryonic antigen-related cell adhesion molecule 4), LAMP1 (lysosomal-associated membrane protein 1), PLA2G7 (phospholipase A2 group VII), and PLAC8 (placenta-specific 8-gene protein) (7). The simultaneous analysis of RNA transcription by these four genes in peripheral blood potentially utilizes information that is contained in various unrelated pathways of the host response at the transcriptome level. This new technology was recently approved by the American Food and Drug Administration. In two technical validation studies SeptiCyte LAB was highly specific for infection in selected groups of both adult and pediatric patients, including some subjects for whom presence or absence of infection was already self-evident at the time of testing (7, 8). As a result, the precise clinical utility of the test for discriminating infectious and non-infectious causes of inflammation in the ICU remains unknown.

Patients for whom a diagnostic biomarker for infection is particularly relevant are those admitted to the intensive care unit (ICU) with acute respiratory failure (ARF) after a previous stay in hospital wards. They frequently suffer from prolonged ICU stays and high mortality (9), yet dyspnea in these patients is a very non-specific symptom and its differential diagnosis is thus extensive, including congestive heart failure, pleural effusion, atelectasis, pulmonary embolus, acute respiratory distress syndrome, and —virtually always— infection. Early confirmation of infection allows timely initiation of antimicrobial therapy, whereas early rejection might prompt a comprehensive diagnostic work-up for non-infectious causes of respiratory distress. Therefore, we aimed to determine the diagnostic and prognostic value of SeptiCyte LAB in hospitalized patients admitted to the ICU with ARF.

Materials and Methods

Study design

For this nested cohort analysis, we selected patients who were enrolled in the Molecular Diagnosis and Risk Stratification of Sepsis (MARS) project, a prospective observational cohort study in the tertiary mixed ICUs of the University Medical Center Utrecht and the Academic Medical Center Amsterdam in the Netherlands (10). Ethical approval for the study was provided by the Medical Ethics Committees of both hospitals, and an opt-out procedure to obtain consent from eligible patients was in place (protocol number 10-056).

Patients

Patients were included if they had been admitted to the ICU between January 2011 and December 2013 with ARF (evidenced by a need for mechanical ventilation within 24 hours of presentation) following prior hospital stay(on a general ward, coronary care unit, or medium care unit) of at least 48 hours. Furthermore, all patients had to have an early warning score >5 (a clinical screening tool based on 6 cardinal vital signs (11)) and/or presence of ≥2 systemic inflammatory response syndrome criteria at ICU admission. Patients were excluded if they had another pertinent need for intubation and ARF was evidently not caused by an infection (including, but not limited to, chronic respiratory insufficiency, primary cardiac arrest, and airway obstruction) or if a diagnosis of infection already had been established at the time of ICU admission (i.e., confirmed infections for which antimicrobial therapy had been started >2 days prior to ICU admission) as the SeptiCyte LAB test was considered to offer little added value in clinical decision making in such patients.

Reference diagnosis

Infectious events were registered upon each occasion that antimicrobial therapy was initiated, and subsequently adjudicated using detailed definitions derived from Center of Disease Control (CDC) and International Sepsis Forum (ISF) Consensus Conference criteria (10, 12, 13). To this end, dedicated physicians not involved in patient care categorized infection plausibility as none, possible, probable, or definite, based on a comprehensive post-hoc review of available clinical, microbiological, and radiological data. Daily discussions between observers and the attending team served to reach consensus in case of any uncertainties. For use as reference test in the current analysis, we reclassified all plausibility ratings into the following categories: infection ruled-out (patients with a post-hoc likelihood rated none, or patients who were not treated for infection), infection undetermined (patients with possible infection), or infection confirmed (patients with a post-hoc likelihood rated probable or definite).

SeptiCyte LAB

Blood specimens were collected within 24 hours of ICU admission in all patients using 2.5 mL PAXgene Blood RNA Tubes (PreAnalytiX GmbH, Hombrechtikon, Switzerland). Samples were kept for a period of 2 to 72 hours at room temperature, and subsequently stored at -20°C (for a maximum of 1 month) and finally stocked at -80°C until analysis. RNA was then isolated on a QIAcube workstation using a PAXgene blood miRNA kit (Qiagen, Venlo, the Netherlands). The concentration of total RNA per sample was assessed by Nanodrop spectrophotometry (Agilent, Amstelveen, the Netherlands) and had to be between 2 and 50 ng/uL to be eligible for further analysis.

SeptiCyte LAB tests were performed in 96-well microtiter amplification plates on an Applied Biosystems 7500 Fast Dx Real-Time PCR Instrument (Thermo Fisher Scientific, Carlsbad, CA). During each amplification run, 3 control samples were included. PCR results were initially quantified using ABI Sequence Detection Software version 1.4. The SeptiCyte LAB score was then calculated from the threshold cycle numbers (Ct-values) measured per gene as follows: *SeptiCyte LAB score* = $(Ct_{PLA2G7} + Ct_{CEACAM4}) - (Ct_{PLAC8} + Ct_{LAMP1})$. The resulting score was finally classified into 4 probability bands reflecting an increasing sepsis likelihood according to the manufacturer's specification; scores ≤3.1 represented band 1 and were categorized as 'sepsis unlikely', whereas scores 3.1-4, 4-6, and >6 represented bands 2, 3, and 4, respectively, and were categorized as 'sepsis likely'.

Statistical analysis

We performed mainly descriptive analyses to determine the diagnostic value of SeptiCyte LAB as formal assessment of test characteristics was precluded due to the large proportion of patients in whom infection status remained inconclusive. However, some diagnostic measures were calculated, not taking into account these latter patients. Furthermore, we calculated the area under the receiver-operating curve (AUROC) to compare the performance of SeptiCyte LAB with CRP, a biomarker commonly used in clinical practice. CRP was not measured at ICU admission in 115 (25%) cases, these values were replaced with estimates derived from multiple imputation (details can be found in Appendix I) (14, 15).

To assess the potential utility of SeptiCyte LAB for risk stratification of patients upon ICU admission, we studied the relation of test results with case fatality (after correction for disease severity). We constructed two prognostic models, using the APACHE IV score either alone or combined with the SeptiCyte LAB score to predict 30-day mortality. We used generalized linear mixed models with a binomial distribution and logit link, and added a random intercept to accommodate possible outcome differences between participating hospitals. Model evaluation was based on Akaike's information criterion (AIC) and the AUROC.

Differences between subgroups of patients were tested using the Wilcoxon Rank-Sum test or the Chi-square test, as appropriate. To test differences in patient characteristics associated with increasing SeptiCyte LAB scores, p-values for trend were calculated using the Cochran-Armitage trend test for dichotomous variables, or one-way ANOVA for continuous variables. If ANOVA suggested a significant association, linear regression with the SeptiCyte probability band as group determinant was performed. All analyses were performed in SAS Enterprise Guide 4.3 (SAS Institute, Cary, NC) and R Studio (R Studio Team 2015, Boston, MA).

Results

Patients

Among 1399 hospitalized patients admitted to the ICU with ARF during the study period, 638 subjects were eligible for inclusion (Figure 1). Blood samples were unavailable in 157 of these, mostly because specimens had been used for prior studies within the MARS-project. Fourteen other patients could not be evaluated due to technical issues during sample preparation or processing, leaving 467 (73%) subjects for final analysis.

Compared to included patients, patients without samples for analysis had less congestive heart failure, more chronic cardiovascular insufficiency, and higher APACHE IV scores and ICU mortality (Appendix II, eTable 1).

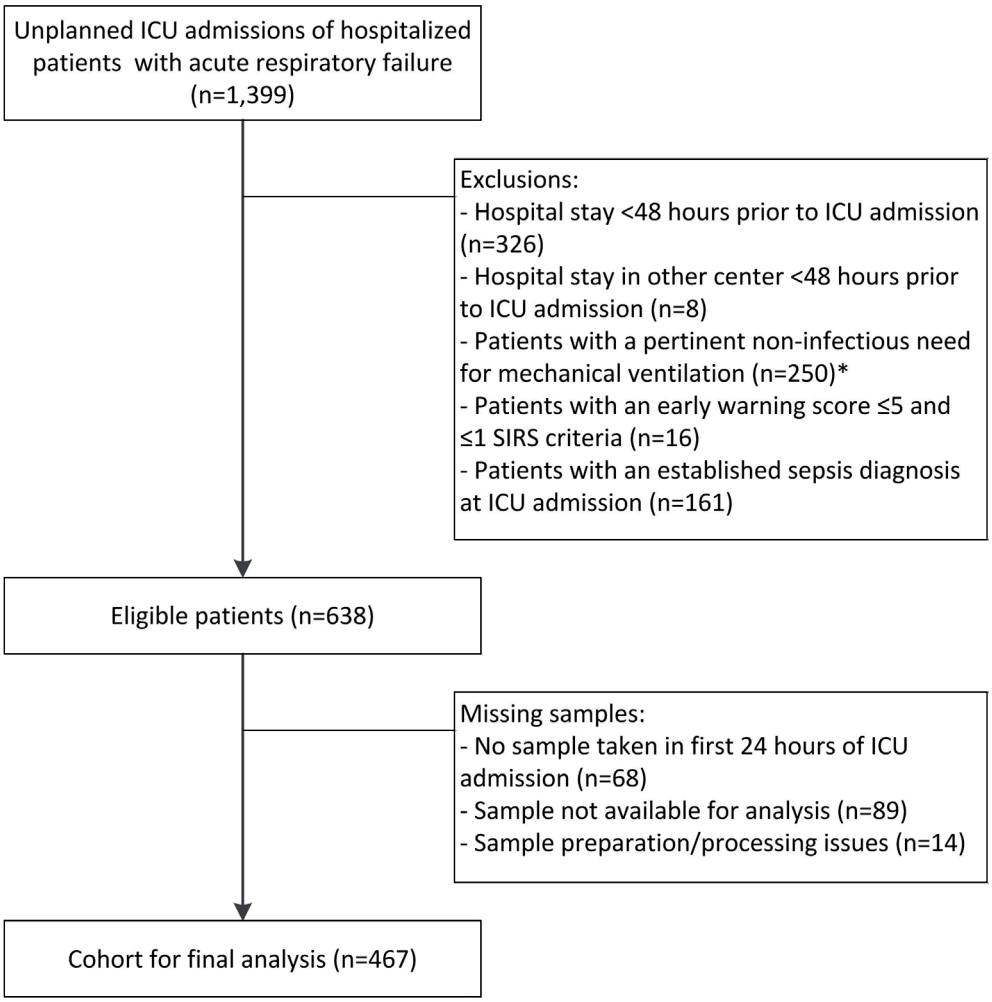
Presence of infection

Because of presumed infection, 359 (77%) of 467 included patients received antimicrobial therapy on day 1 in the ICU, and another 14 subjects (3%) started treatment on day 2. Among these, the post-hoc plausibility of infection was rated none in 41 (11%) cases. An additional 94 patients did not receive antimicrobial therapy during the first 2 days in the ICU, yielding a total of 135 subjects in whom infection was considered ruled-out. The remaining 332 patients were classified as undetermined (n=135) or infection confirmed (n=197). Hence, in the total study population, the pre-test probability of infection was 197/467 (42%). Of the patients in whom infection was undetermined or confirmed, the most commonly suspected sites of infection were respiratory tract infections (n=228), abdominal infections (n=52), and bloodstream infections (n=36).

SeptiCyte LAB results

In patients in whom infection was ruled-out (n=135), undetermined (n=135), or confirmed (n=197), median (IQR) SeptiCyte LAB scores were 4.8 (3.7-6.1), 5.3 (3.9-6.4), and 6.5 (5.2-8.1), respectively (Figure 2).

Figure 1. Flowchart of patient inclusions

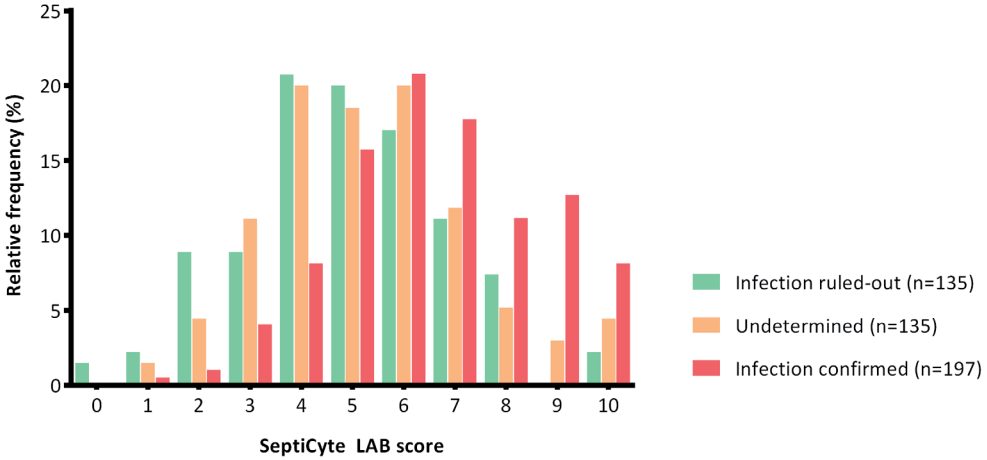


ICU: intensive care unit. SIRS: systemic inflammatory response syndrome. *Including, but not limited to, patients with chronic respiratory insufficiency (n=107), in-hospital cardiac arrest (n=103), and airway obstruction (n=18).

Formal analysis yielded a significant correlation between test scores and the probability of infection (Spearman's rho 0.320; p<0.001). However, Ct-values for all four individual genes were largely overlapping (Appendix III, eFigure 1).

Table 1 shows the probability bands for infection at admission according to the SeptiCyte LAB score in relation to the reference diagnosis. Dichotomizing test results at band ≥2 (as per manufacturer specification (7)), concordance was observed in 189 (96%) of 197 patients with confirmed infection, yet in only 25 (18%) of 135 patients in whom infection was eventually ruled-out. Using SeptiCyte LAB at this cut-off to select patients for antimicrobial treatment would have led to inappropriate prescriptions in

Figure 2. Distribution of SeptiCyte LAB scores by reference diagnosis



A higher SeptiCyte LAB score indicates a higher likelihood of sepsis. A score ≤3.1 should be interpreted as sepsis unlikely according to the manufacturer's specification.

Table 1. SeptiCyte LAB result versus reference diagnosis

Probability band[a]	Infection ruled-out (n=135)	Undetermined (n=135)	Infection confirmed (n=197)
Band 1: Sepsis unlikely (n=52)	25 (19)	19 (14)	8 (4)
Band 2: Sepsis likely (n=39)	17 (13)	15 (11)	7 (4)
Band 3: Sepsis likely (n=181)	57 (42)	57 (42)	67 (34)
Band 4: Sepsis likely (n=195)	36 (27)	44 (33)	115 (58)

Data presented as n (%). All percentages are column percentages. [a] Higher SeptiCyte probability bands indicate increased likelihood of sepsis.

110 cases (of which only 38 were currently treated). After exclusion of undetermined cases, the positive predictive values for probability bands 2, 3, and 4 were 29%, 54%, and 76%, respectively, and the negative predictive value for probability band 1 was 76%.

Discrepancy analysis

We observed 8 discordant cases where the test suggested that infection could be safely ruled-out, whereas infection was confirmed on post-hoc assessment. Review of these false negative cases revealed that these patients were older, had lower severity of illness upon presentation to the ICU, and more frequently had been previously admitted to the ICU than the 189 patients with true positive results (Table 2).

Table 2. Characteristics of patients according to test result (discrepancy analysis)

	Infection ruled-out (n=135)		Infection confirmed (n=197)	
Patient and ICU characteristics	**True negative**	**False positive**	**True positive**	**False negative***
Patients, N	25	110	189	8
Gender, female	14 (56)	48 (44)	58 (31)	1 (13)
Age, years	60 (48-67)	64 (53-75)	65 (56-73)	72 (64-77)
Surgical reason for admission	1 (4)	9 (8)	9 (5)	1 (13)
Comorbidities				
- Congestive heart failure	3 (12)	10 (9)	10 (5)	0 (0)
- COPD	3 (12)	8 (7)	23 (12)	2 (25)
- Chronic cardiovascular insufficiency	3 (12)	1 (1)	6 (3)	1 (13)
- Diabetes mellitus	4 (16)	24 (22)	36 (19)	2 (25)
Immune deficiency	3 (12)	16 (15)	30 (16)	2 (25)
APACHE IV Score	73 (60-86)	81 (64-100)	84 (70-101)	64 (61-103)
Suspected site of infection				
- Respiratory tract	1 (4)	20 (18)	93 (49)	6 (75)
- Abdominal	0 (0)	3 (3)	40 (21)	1 (13)
- Cardiovascular	1 (4)	3 (3)	24 (13)	1 (13)
- Other/unknown	1 (4)	12 (11)	32 (17)	0 (0)
- No suspicion	22 (88)	72 (65)	0 (0)	0 (0)
Prior ICU admission during hospital stay	5 (20)	57 (52)	92 (49)	6 (75)
ICU length of stay, days	2 (1-8)	4 (2-8)	7 (3-12)	4 (2-8)
ICU mortality	2 (8)	18 (16)	36 (19)	0 (0)

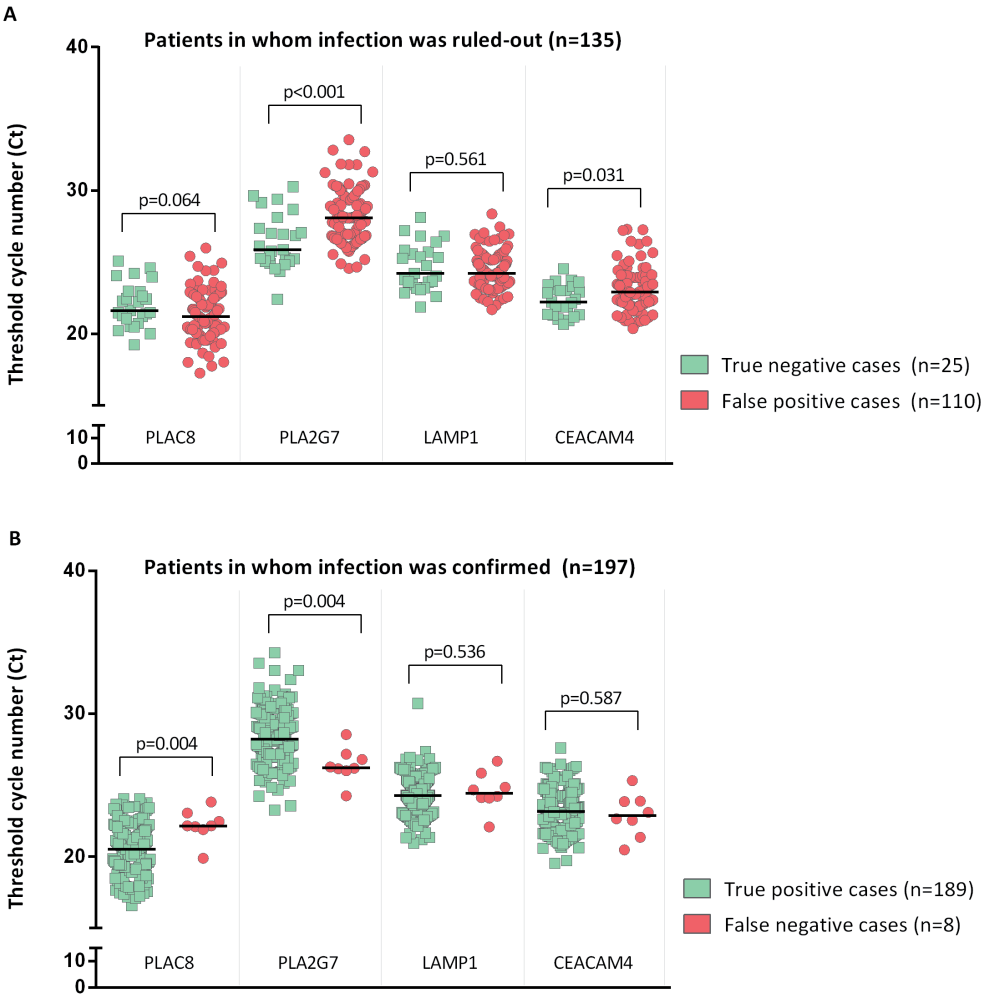
COPD: chronic obstructive pulmonary disease. CHF: congestive heart failure. APAHCE: acute physiology and chronic health evaluation. ICU: intensive care unit. Continuous data are presented as medians (IQR), dichotomous data are presented as n (%). *Case descriptions for these patients are provided in Appendix III.

Case descriptions for these patients are provided in Appendix IV. In-depth analysis of the 110 patients with false positive results revealed that they had similar age, higher severity of illness, more previous ICU admissions, and were more likely to have been clinically suspected of infection than their 25 true negative counterparts. In addition, we compared individual RNA transcripts in discrepant cases. Ct-values differed significantly for the PLA2G7, CEACAM4 and (possibly) PLAC8 genes, but not for LAMP1, when comparing false positive to true negative results (Figure 3A). Similarly, there were significant differences in the median Ct-values of PLAC8 and PLA2G7 when comparing false negative and true positive test results, but not for LAMP1 and CEACAM4 (Figure 3B).

Comparative diagnostic evaluation

To better assess the clinical utility of SeptiCyte LAB in the ARF population, we compared its diagnostic performance to CRP. In patients in whom infection was ruled-out, undetermined, or confirmed, median (IQR) plasma concentrations of CRP at ICU admission were 67 (22-152), 109 (63-207), and 166 (93-252) mmol/L, respectively. After

Figure 3. Median Ct-values per gene for non-infectious and infectious cases according to test result



For this analysis, cases in which infection was undetermined (n=135) were not taken into account as it was unknown whether the test classified them correctly.

exclusion of undetermined cases, ROC analysis yielded an AUC of 0.727 (95%CI 0.666-0.788) for CRP versus 0.731 (95%CI 0.677-0.786) for SeptiCyte LAB (mean difference 0.004, 95%CI −0.077-0.086; p=0.919).

Prognostic evaluation

Higher SeptiCyte LAB scores were associated with both greater disease severity upon ICU admission and increased mortality (Appendix V, eTable 2). However, a prognostic model that included both APACHE IV and SeptiCyte LAB scores was not superior in

predicting 30-day mortality compared to a model using only the APACHE IV score (AUROC 0.737 versus 0.735, p=0.724; AIC 498 versus 497).

Discussion

In this study, we evaluated the clinical utility of SeptiCyte LAB to diagnose infection in patients admitted to the ICU because of ARF after a period of hospitalization. Infectious episodes were correctly identified by the test in 96% of the patients with confirmed infection. However, in patients in whom an infection was refuted, the test yielded a correct result in only 18% of patients. In fact, in this population the test did not offer better diagnostic discrimination than the more commonly used biomarker CRP. In addition, SeptiCyte LAB did not improve prognostication when added to the APACHE IV score.

Previous studies of SeptiCyte LAB reported very high discriminative power for infection (AUCs of 0.88 and 0.99) compared to what we observed (7, 8). The major difference between those studies and ours concerns the study domain. Early validation studies have mostly used cohorts in which infectious and non-infectious patients could clearly be distinguished on clinical grounds. For instance, one study compared children after cardio-pulmonary bypass surgery to children with severe sepsis (8). In another preliminary evaluation of SeptiCyte LAB (which included 23 subjects also enrolled in the current study), the test performed better in a cohort of highly selected patients (AUC 0.95; 95%CI 0.91-1.00) than in a cohort representing a more real-life setting (AUC 0.85; 95%CI 0.75-0.95) (7). Furthermore, an assessment of its diagnostic performance across 39 publicly available datasets yielded highly variable findings, with reported AUCs ranging from 0.24 to 1 for individual datasets (16). In search of a possible explanation for the lack of discriminative performance of SeptiCyte LAB in certain subgroups (some of which did not represent the intended use population of the test), it was noted that the expression of one of the four genes involved in calculating the SeptiCyte LAB score (CEACAM4) was down regulated during sepsis in the discovery cohort, but not in other cohorts (16). In our study, we observed only minimal differences in gene expression between infectious and non-infectious cases for all four genes, including CEACAM4 (Appendix III, eFigure 1).

We deliberately focused on a target population in which it was difficult to diagnose infections with certainty. Many patients had significant (acute) comorbidities, had stayed in the hospital for prolonged periods of time prior to ICU admission, and had previously been exposed to antimicrobials for (presumed) infections. To avoid selection, we enrolled consecutive patients. However, 171 of 638 eligible patients were excluded from analysis, mostly because they had a clinically apparent sepsis syndrome (due to confirmed infection) and their samples had already been used for other studies within

the MARS-project. These exclusions thus enriched our study cohort with infectious episodes that were more challenging to diagnose. Yet, we believe there is little value in using SeptiCyte LAB (or any other biomarker) in patients with clinically overt infection.

Although the probability of infection was prospectively adjudicated by trained observers based on available post-hoc clinical, radiological, and microbiological findings, some diagnostic misclassification will most likely have occurred (10). For instance, infectious episodes for which treating physicians did not initiate antimicrobial therapy may have been erroneously classified as infection ruled-out. It is important to stress that, in the presence of an imperfect reference test, the maximal discriminative performance (in terms of AUROC) that can be attained by a diagnostic test will be necessarily lower than 1. Thus, diagnostic misclassification may have reduced the apparent diagnostic utility of SeptiCyte LAB in our cohort. This merely emphasizes the difficulty of performing diagnostic studies in patients with infection, where a gold standard simply does not exist. However, it is unlikely that the lack of a robust reference diagnosis explains the observed differences in discriminative power of the test between our study and previous validation cohorts, nor the equipoise between SeptiCyte LAB and a more common host-response marker such as CRP in classifying infections (7).

Conclusions

In our clinical evaluation of SeptiCyte LAB in patients presenting to the ICU with ARF after prior hospitalization for other acute diseases, the discriminative power of this new biomarker test was lower than previously reported in more selective validation cohorts. As SeptiCyte LAB scores are based on gene expression profiles, test results might vary between specific populations and/or settings. Therefore, more prospective studies are needed to determine the clinical utility of this novel test.

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SUPPLEMENTARY MATERIAL

Appendix I: Multiple imputation model and handling of imputed data

Multiple imputation model

CRP was not measured at ICU admission in 115 (25%) of 467 cases. Since complete case analysis in this situation may lead to bias (1), we chose to perform multiple imputation. The use of multiple imputed datasets accounts for uncertainty introduced by imputation, providing larger standard errors when the number of missing values increases. To impute CRP values that were missing on the first day in the ICU we used an imputation method based on chained equations (R-package 'mice', version 2.25, 2015) (2-3). In this we assumed data were missing at random (i.e., that any systematic differences between missing and observed values could be explained by other parameters in our data set) (4). Variables that were used in the imputation model to predict missing CRP values included: gender, age, race, medical or surgical admission, immune deficiency, Charlson comorbidity index, APACHE IV score, SOFA score at ICU admission, presence of infection at admission, SIRS criteria (fever, tachycardia, tachypnea, and abnormal white blood cell count) at ICU admission, ICU length of stay, and the SeptiCyte LAB score.

If needed, rounding and boundaries of imputed values were used to assure that clinically possible values were replaced for missing values (5). Considering that 25% of cases had missing values for CRP, we performed 25 imputations with 30 iterations per imputation. This resulted in stable imputations as evidenced by summary statistics and density plots (3, 6-8).

Handling of imputed data

We averaged AUROCs as estimated from the 25 separate datasets in order to arrive at a robust estimate for the discriminative power of CRP. Also, the difference in AUCs was calculated as the average difference between the AUROCs of SeptiCyte and CRP across all imputed datasets. Using Rubin's rules, we calculated the accompanying 95% confidence intervals and test-statistics for these estimations to arrive at correct effect estimates and standard errors (9-10).

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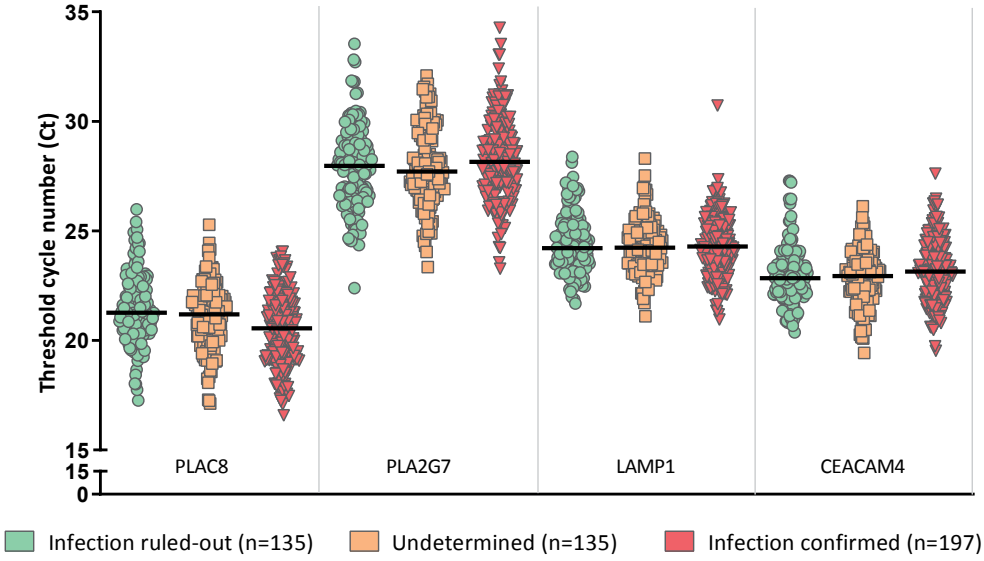
Appendix II: eTable 1. Characteristics of in- and excluded patients

Patient characteristics	Exclusions (patients without sample and/or test result, n=171)	Inclusions (patients with test result, n=467)	P-value
Gender, female	66 (39)	169 (36)	0.577
Age, years	65 (54-73)	65 (54-73)	0.870
Surgical reason for admission	6 (4)	25 (5)	0.337
Comorbidities			
- Congestive heart failure	4 (2)	33 (7)	0.024
- COPD	20 (12)	52 (11)	0.843
- Chronic cardiovascular insufficiency	13 (8)	17 (4)	0.036
- Diabetes mellitus	24 (14)	88 (19)	0.157
Immune deficiency	33 (19)	68 (15)	0.147
APACHE IV Score	87 (70-111)	81 (65-98)	0.010
Admission diagnosis (top 3 categories)			0.098
- Pneumonia	38 (22)	131 (28)	
- Congestive heart failure	13 (8)	34 (7)	
- Atelectasis	12 (7)	32 (7)	
Prior ICU admission during hospital stay	75 (44)	227 (49)	0.287
Intubation	137 (80)	394 (84)	0.203
ICU length of stay, days	4 (2-11)	5 (2-10)	0.104
ICU mortality	42 (25)	74 (16)	0.012

COPD: chronic obstructive pulmonary disease. CHF: congestive heart failure. APACHE: acute physiology and chronic health evaluation. ICU: intensive care unit. Continuous data are presented as medians (IQR), dichotomous data are presented as n (%).

Appendix III: eFigure 1. Ct-values per gene in the SeptiCyte LAB test per reference category



The vertical black lines indicate the median Ct-value for that reference category.

Appendix IV: Case vignettes of false negative cases

Patient #1
Patient had been previously admitted to ICU because of bilateral pneumonia. After 2 weeks she was readmitted with respiratory insufficiency mainly due to fluid overload and atelectasis. However, the patient was also suspected of recurrent pneumonia and because of earlier growth of Enterobacter cloacae, she was empirically started on vancomycin and ciprofloxacin. Sputum cultures taken at admission subsequently grew Staphylococcus aureus, after which treatment was switched to flucloxacillin. Blood cultures remained negative.

Patient #2
Patient had been treated in the ICU for mediastinitis following a spontaneous retropharyngeal abscess for 6 weeks. Five days after discontinuation of antimicrobial therapy he was readmitted with tachypnea and fever. CRP 31/93. The differential diagnosis on admission included HAP, pulmonary embolism, recurrent abscess, and empyema. Empirical treatment with ceftriaxone plus metronidazole was initiated. CT-guided percutaneous drainage of evident mediastinal pus collections was performed two days after admission, cultures of which remained negative.

Patient #3
Patient had been admitted to ICU for 5 days following esophagectomy. After 3 months he was readmitted with dyspnea, fever and back pain. CRP 79/193. PCT 2/4. The differential diagnosis on admission included HAP, pulmonary embolism, and late anastomotic leakage. Chest X-ray showed both pulmonary infiltrates and spinal fractures (due to metastatic disease). Blood cultures taken at admission grew Klebsiella pneumonia and Klebsiella oxytoca. The patient was treated with ceftriaxone.

Patient #4
History of DM induced nephropathy, for which continuous ambulatory peritoneal dialysis. Patient had been treated for 3 months with intraperitoneal vancomycin for catheter peritonitis (due to corynebacterium) until 4 months earlier, and had undergone CABG 1 month before ICU admission. He was admitted with shock, respiratory insufficiency and left-sided pleural effusion. CRP 234. The differential diagnosis on admission included pneumosepsis, abdominal sepsis (due to recurring CAPD peritonitis), bowel ischemia, and hematothorax. An explorative laparotomy was performed, but was negative. Sputum cultures taken on the day of admission grew Klebsiealla pneumoniae and Serratia marcescens. The patient recovered following ciprofloxacin treatment.

Patient #5

Patient had been admitted to ICU for 5 days following pancreatectomy. After 3 weeks he was readmitted with hypothermia, lactic acidosis and hypercapnia. CRP 55. The differential diagnosis on admission included anastomotic leakage, abdominal abscess, pleural effusion, and exacerbation of COPD. Laparotomy showed small bowel perforation with an infected pocket in the right upper quadrant, pus cultured from the pocket grew a small amount of Candida albicans. Patient was empirically treated with ciprofloxacin, clindamycin, and anidulafungin.

Patient #6

History of laryngectomy. Patient underwent uncomplicated mandibular resection. Three days later he was admitted to the ICU with hypoxia and (some) fever (38.2). CRP 76/155. PCT 0/0. The diagnosis was evident: massive aspiration pneumonia due to malfunction of his pre-existing (one-way) tracheal-oesophageal speaking valve. He was empirically started on ceftriaxone and therapy was completed before the final results of the sputum cultures, taken on the day of admission, were known. These cultures grew Enterobacter cloacae and Pseudomonas aeruginosa.

Patient #7

Immunocompromised patient following allogenic stem cell transplant for CML. Medical history of recurrent infections, most currently cellulitis of the left lower leg for which he was still receiving flucloxacillin. Presentation to ICU with hematemesis (most likely due to GvHD) and a new pulmonary infiltrate. Patient was treated empirically with ceftazidime, clindamycin and voriconazole. Neither BAL nor blood cultures (which were both performed while patient was receiving antimicrobial treatment) yielded a probable causative pathogen.

Patient #8

Patient had been admitted to ICU for 4 days following acute subdural hematoma (while on anticoagulation) for which surgical decompression had been performed. Ten days later he was readmitted with fever, new-onset atrial fibrillation, renal insufficiency, and respiratory distress. CRP 112/185. The differential diagnosis on admission included aspiration pneumonia (due to difficulty swallowing), wound infection (he had a lesion on the back of his head), secondary meningitis , and decompensated heart failure. Patient was empirically treated with ceftriaxone (for possible pneumonia) and flucloxacillin (for presumed wound infection), but later switched to ciprofloxacin when the sputum and blood cultures taken at admission both grew Serratia marcescens.

Appendix V: eTable 2. Patient characteristics by SeptiCyte LAB result

	SeptiCyte probability band				
Patient characteristics	**Band 1 (n=52)**	**Band 2 (n=39)**	**Band 3 (n=181)**	**Band 4 (n=195)**	**P-value for trend**
Gender, female	21 (40)	13 (33)	67 (37)	68 (35)	0.562
Age	67 (54-76)	65 (53-75)	63 (53-73)	65 (55-73)	0.710
Immune deficiency	7 (13)	5 (13)	21 (12)	35 (18)	0.253
APACHE IV score	74 (63-96)	69 (59-89)	80 (64-96)	84 (70-102)	<0.001
Prior ICU admission during hospital stay	17 (33)	22 (56)	97 (54)	91 (47)	0.352
ICU length of stay, days	5 (2-9)	4 (2-8)	6 (3-11)	6 (3-10)	0.306
ICU mortality	4 (8)	6 (15)	24 (13)	40 (21)	0.020
30-day mortality	7 (14)	11 (28)	37 (20)	46 (24)	0.271

Continuous data are presented as medians (IQR), dichotomous data are presented as n (%).

Addendum to Chapter 2

Author correspondence

Maria E. Koster-Brouwer
Diana M. Verboom
Marc J.M. Bonten
Olaf L. Cremer

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In response to McHugh LC, Davis RF, Yager TD: Uncertainty in diagnosis leads to underestimates of performance, Critical Care Medicine (2018)

The importance of selecting patients with a true diagnostic dilemma in diagnostic research

McHugh *et al.* argue that our diagnostic assessment of SeptiCyte LAB in previously hospitalized patients admitted to the intensive care unit (ICU) with acute respiratory failure may have been negatively affected by the choice of study population (1). Indeed, the study aimed to validate this novel biomarker in a cohort with a true medical need to perform a diagnostic workup for infection as — in real life — a diagnostic test may influence clinical decision making only in such patients, and not in subjects in whom an infection has already been confirmed and treated before ICU admission or in whom there exists an evidently non-infectious reason for respiratory distress. This deliberately differs from previous biomarker studies in sepsis (2, 3). Our cohort exclusively consisted of patients in whom the physician initially was not certain about the presence of infection. Adjudication of infection plausibility (the reference test) was subsequently based on a post-hoc assessment by independent research physicians who were blinded to SeptiCyte LAB results and not involved in patient care. The same classification system was used in a previous evaluation of this test (2). Therefore, the (lack of) confidence by which a physician decided to start or withhold therapy at the time of ICU presentation cannot be equated to misclassification error in our reference test, which explicitly acknowledges residual diagnostic uncertainty by including a separate category for 'undetermined' cases (*n*=135/467). To be fully transparent we reported these subjects yet did not include their data when calculating performance measures of SeptiCyte LAB, thereby potentially over- rather than underestimating test performance.

The second critique point refers to the use of CRP to diagnose infection. As explained in the manuscript, definitions of confirmed infection in our study relied completely on clinical, radiological and microbiological evidence, and CRP was thus not part of any of these. Solely in cases of hospital-acquired pneumonia —and in the absence of any (other) clinical or inflammatory symptoms— a CRP level >30 mmol/L could have resulted in the categorization of a suspected infection as 'undetermined' instead of 'ruled-out'. However, this did not occur in our study cohort. Furthermore, cases in which infectious status remained undetermined were excluded from the direct comparison between both biomarkers. We therefore infer that the availability of CRP to assessors did not influence categorizing infection plausibility and thus did not impact the study results.

In conclusion, we contend that our observations accurately reflect the diagnostic performance of SeptiCyte LAB in patients in whom a true diagnostic dilemma is present.

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Lieve tante Marjan,
Vanharte beterschap
hoop dat u snel
weer opknapt
liefs Johan. Heleen
en de kids

Part II

Estimating economic burden

3

Chapter 3

Critical care management of severe sepsis and septic shock: a cost-analysis

Maria E. Koster-Brouwer
Peter M.C. Klein Klouwenberg
Wietze Pasma
Judith E. Bosmans
Tom van der Poll
Marc J.M. Bonten
Olaf L. Cremer
On behalf of the MARS consortium

Published in Netherlands Journal of Critical Care (2016)

Abstract

Background Sepsis treatment has been associated with high costs. Furthermore, both the incidence of sepsis and the severity of illness at presentation appear to be increasing. We estimated health care costs related to the treatment of patients with sepsis in the intensive care unit (ICU) and aimed to explain variability in costs between individuals.

Methods We performed a prospective cohort study in patients presenting to the ICU with severe sepsis or septic shock to two tertiary centres in the Netherlands. Resource use was valued using a bottom-up micro-costing approach. Multivariable regression analysis was used to study variability in costs.

Results Overall, 651 patients were included, of which 294 presented with septic shock. Mean costs were €2,250 (95%CI €2,235-€2,266) per day and €29,102 (95%CI €26,598-€31,690) per ICU admission. Of the total expenditure, 74% was related to accommodation, personnel, and disposables, 12% to diagnostic procedures, and 14% to therapeutic interventions. Patients with septic shock had higher costs compared to patients with severe sepsis (additional costs: €69 (95%CI €37-€100) per day, and €8,355 (95%CI €3,400-€13,367) per admission). Site of infection, causative organism, presence of shock, and immunodeficiency were independently associated with costs, but explained only 11% of the total variance.

Conclusion Mean costs of sepsis care in the ICU were almost €30,000 per case. As costs were poorly predictable, opportunities for costs savings based on patient profiling upon admission are limited.

Introduction

Over the last decades, both the reported incidence of sepsis in the general population and the severity of disease at presentation to the hospital are rising (1, 2). These increases may be explained by an aging population that brings about more chronic comorbidities. Alternatively, raised awareness amongst doctors as a result of the Surviving Sepsis Campaigns may have resulted in an overdiagnosis of sepsis to some extent (1, 2). Sepsis patients who develop organ failure are considered to have severe sepsis, whereas those with advanced circulatory compromise are considered to be in septic shock (3). In either case, management in a critical care facility is required. As the technological possibilities to provide effective life support to critically ill patients have advanced, both the hopes of individual patients as well as the general public demand to have access to the 'best' possible care have fostered. Hence, doctors in intensive care units (ICU) are faced with increasingly difficult decisions to stop or deny further treatment. Taken together, these developments will likely result in both a higher demand for sepsis treatment in the ICU, as well as increased expenditures per patient (1, 4, 5).

Insight into the costs of sepsis care and its determinants, is of major importance in this era of universal restraints on health care budgets. Several authors have estimated expenditures for sepsis treatment in different settings and from different perspectives, including estimations of total hospitalization costs for sepsis and costs of sepsis in developing countries (4-9). However, only very few studies have specifically investigated costs of sepsis treatment in ICU settings (5, 6, 9). Moreover, available studies predominantly relied on administrative data to identify sepsis cases and did not use individual patient data to generate cost estimates. These methods do not adequately reflect the variation in costs between patients, precluding the possibility to explain individual variability by multivariable modelling (10). Furthermore, these studies used data that were collected during the 1990s, and may thus no longer be representative of the current health care situation.

Although the existing literature is somewhat inconsistent, several characteristics are believed to be (independently) associated with high costs of treatment, including advanced age (5, 11), increased severity of illness (5), a surgical reason for admission (5, 6, 11), and some specific sites of infection (especially catheter-related infections) (9). Knowledge about patient and illness characteristics that are associated with excessive health care costs might be helpful for clinicians to make informed decisions when considering treatment of critically ill patients with sepsis.

In this study, we estimated the total direct health care costs associated with treatment of patients with severe sepsis or septic shock in the ICU using a bottom-up micro-costing approach. Furthermore, we investigated to what extent patient and illness characteristics measured at baseline can explain variability in costs between patients.

Methods

Design

Data were collected within the Molecular Diagnosis and Risk Stratification of Sepsis (MARS) project, a prospective cohort study in two tertiary ICUs in the Netherlands (12). Ethical approval for the study was provided by the Medical Ethics Committee of the University Medical Center Utrecht. They also gave permission for an opt-out procedure to obtain consent from eligible patients (IRB number 10-056).

Participants

We included consecutive adults presenting to the ICUs of the University Medical Center Utrecht or the Academic Medical Center Amsterdam between January 2011 and October 2013 with severe sepsis or septic shock as their main reason for admission. We used definitions for severe sepsis and septic shock that were compliant with the 2008 Surviving Sepsis Campaign guidelines (3). We only included patients having community-acquired or nosocomial infections with an onset within 72 hours before ICU admittance, and a post-hoc plausibility of infection that was graded as probable or definite by dedicated observers using validated definitions (12). Cases of unconfirmed (possible) sepsis were excluded, as were patients that had been admitted from, or were discharged to another ICU, because data on total resource use and costs in these patients could not be completed.

Data collection

Trained observers prospectively collected data on patient and illness characteristics, including demographics, chronic comorbidities, admission type, Acute Physiology and Chronic Health Evaluation (APACHE) IV score sites of infection, and causative organisms. The sites of infection were classified into five main categories based on their frequency of occurrence: community-acquired pneumonia, hospital-acquired pneumonia, secondary peritonitis, urinary tract infection, bloodstream infection, and other (12). Causative organisms were categorized as Gram-positive bacteria, Gram-negative bacteria, fungi and yeasts, viruses, and unknown.

Utilization of drugs, fluids, nutrition, and blood products was collected using the bedside patient data management system (Metavision, iMDsoft, Israel). Information about the numbers and types of blood chemistry, microbiology, and imaging tests, as well as source control surgical procedures was extracted from the electronic health records of both participating hospitals. In one of the centres information on the number of performed blood chemistry tests was only partially accessible. Although summary test results were available (for example the daily lowest and highest plasma concentration of sodium), it was not possible to derive the exact number of individual blood specimens that these aggregates originated from. Therefore, the costs associated

with blood chemistry for the patients in this centre (enrolling 55% of the total cohort) were estimated following a multivariable imputation procedure based on the complete laboratory data set of patients in the other participating centre.

Costs estimations

We studied direct medical costs during the ICU stay only because we performed our cost-analysis from a health care perspective (rather than from a societal perspective, which would also include elements such as costs for productivity loss and opportunity costs). We categorized resource use into three main categories: accommodation, diagnostic procedures, and therapeutic interventions. Costs for accommodation were derived from a previous study investigating the costs of ICU stay in the Netherlands (13). These comprised expenditures for clinical personnel, general disposables, hotel (including basic dietary costs), overhead (including costs for general expenses, administration, energy, maintenance, insurance, and non-clinical personnel), and capital (including investment costs for buildings and inventory). We identified and valued diagnostic and therapeutic resource use for each patient individually using a bottom-up micro-costing approach. Unified internal tariffs of the participating hospitals were used to value each intervention, test, or procedure. Costs for diagnostic procedures were divided in blood chemistry (which also included immunology and biopsy procedures), microbiology, and imaging. Costs for therapeutic interventions were divided in drugs, fluids, nutrition, blood products, and surgical procedures for source control. In order to calculate total costs per day and per patient, resources use was multiplied by unit price. All costs were expressed in euros and indexed to the 2013 price level.

As we exclusively included patients that had severe sepsis or septic shock as their main reason for admission, we assumed that all costs generated in the ICU were directly sepsis-related. Naturally, many patients had comorbidities, but we considered it not feasible to distinguish costs of sepsis treatment from those resulting from underlying diseases. Nonetheless, we chose to disregard (mainly elective) surgical procedures that were clearly not sepsis-related, because, despite their infrequent occurrence, these events may have a disproportional impact on total costs. All surgery and interventional radiology undertaken as a direct consequence of the sepsis episode (for instance as a source control procedure or to treat complications) were included, however.

Because our study aimed to focus on costs of critical care management exclusively, we chose not to include expenditures during the extended ward stay of patients. However, if patients were re-admitted to the ICU following a primary sepsis event, we accumulated costs over consecutive ICU episodes.

Statistical analysis

Patient characteristics were compared between groups using the chi-square or the Mann-Whitney U test, as appropriate. Costs per day and per accumulated ICU

admission were compared between patients with severe sepsis and septic shock, by different sites of infection, by different causative organisms, and for survivors and non-survivors. Statistical differences in costs between these subgroups were evaluated using the Kruskal-Wallis H test, as appropriate.

We constructed a multivariable linear regression model to explain variability in ICU expenditure between patients. First, univariable linear regression analyses were performed to investigate which patient and disease characteristics were associated with costs. Subsequently, all determinants with a p-value <0.25 in the univariable linear regression analyses were included in a multivariable model. The category with the lowest costs was used as a reference category in these analyses. A manual stepwise backwards regression analysis was then used to select the optimal model to explain variability between patients. The coefficient of determination (R^2) was used to assess the explained variability by the model.

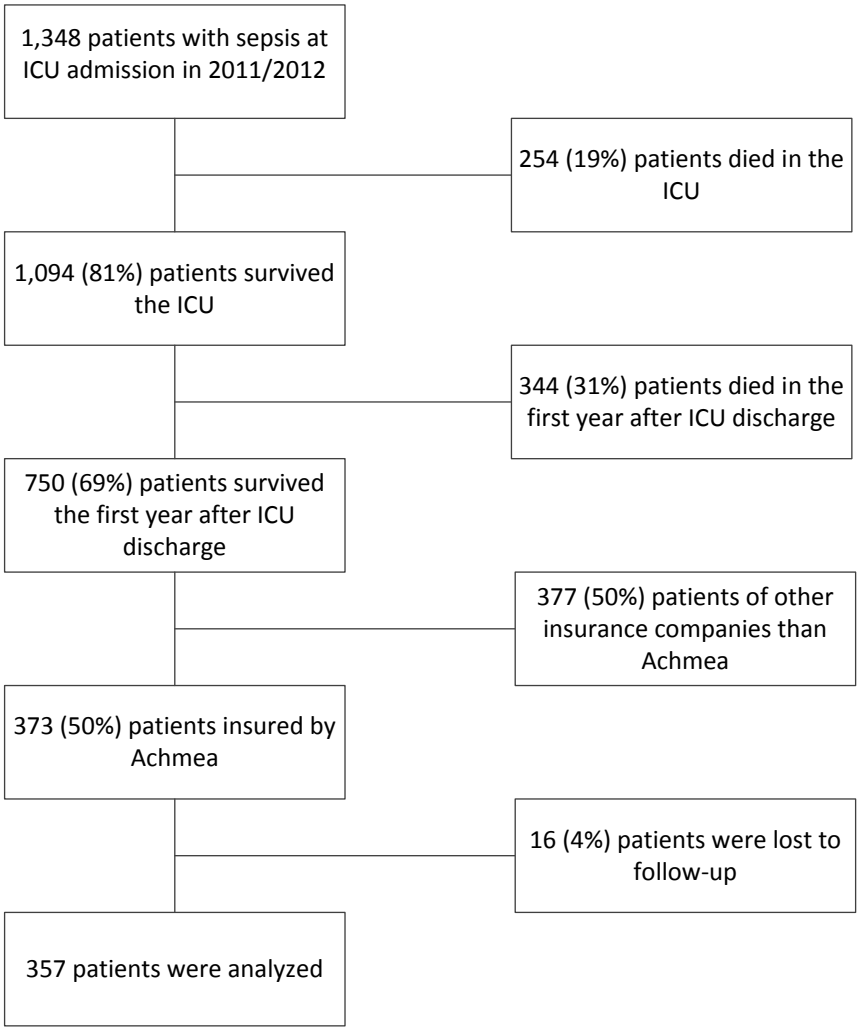
Distributions of costs are typically skewed to the right, due to the presence of few people with very high costs. Because this skewed distribution might result in an overestimation of mean costs, we re-sampled the data 5,000 times with replacement and estimated 95% confidence intervals based on these bootstrapped samples. Bootstrapped confidence intervals do not depend on the assumption that the underlying data follows a normal distribution and are therefore suitable to use in cost studies (14). All analyses were performed in SPSS Version 20.0 for Windows (SPSS, Chicago, IL, USA).

Results

Figure 1 is the flowchart of patient inclusions. In total, we studied 651 patients (contributing 780 ICU episodes in total), of whom 294 (45%) were admitted with severe sepsis and 357 (55%) with septic shock. Patients presenting with septic shock had similar demographic characteristics and comorbidities, but significantly increased markers of acute disease severity and higher mortality compared to patients with severe sepsis (Table 1).

Table 2 shows detailed information on resource use and associated median expenses per day and per admission. The total median costs were €17,659 (IQR €8,623-€37,018) per sepsis episode. However, due to a very skewed distribution of costs, mean costs differed considerably from median costs. Overall, we estimated the mean total costs for treatment of an episode of sepsis in the ICU at €2,250 (95%CI €2,235-€2,266) per day and €29,102 (95%CI €26,598-€31,690) per admission. Accommodation represented 74% (€21,654, 95%CI €19,772-€23,445) of these costs, whereas diagnostic procedures accounted for 12% (€3,457, 95%CI €3,202-€3,726) and therapeutic interventions for 14% (€3,991, 95%CI €3,529-€4,487) of the mean total costs per admission. Costs for diagnostic procedures consisted mainly of expenses for laboratory tests. Although

Figure 1. Flowchart of patient inclusion



ICU: intensive care unit. 36 patients (12%) admitted with severe sepsis progressed to septic shock during ICU admission. For all analyses, patients stayed in the admission category.

the price for several advanced radiological investigations was considerable, these procedures contributed only little to the total diagnostic expenditure because they were utilized in less than 25% of admissions. Drugs represented the largest component of the costs associated with therapeutic interventions, although antibiotics accounted only for a small part of these. However, the costs of antimicrobial therapy differed largely between patients. Similar to what was the case for advanced imaging. Also source control procedures were expensive on a case-by-case basis, but contributed little to overall median costs because they were only performed in a minority of patients.

Table 1. Patient and illness characteristics stratified by sepsis severity

	Severe sepsis (n=294)	Septic shock (n=357)	Total (n=651)	P-value
Gender, male	160 (54)	212 (59)	372 (57)	0.20
Age, years	62 (52-71)	63 (53-71)	63 (53-71)	0.64
Comorbidities				
- Cardiovascular disease[a]	88 (30)	117 (33)	205 (32)	0.45
- Chronic obstructive pulmonary disease	38 (13)	54 (15)	92 (14)	0.43
- Diabetes mellitus	60 (20)	71 (20)	131 (20)	0.92
- Immune deficiency[b]	75 (26)	95 (27)	170 (26)	0.79
Surgical admissions	64 (22)	96 (27)	160 (25)	0.14
APACHE IV score	81 (64-98)	94 (77-118)	87 (71-112)	<0.001
Site of infection				
- Community-acquired pneumonia	88 (30)	77 (22)	165 (25)	0.02
- Hospital-acquired pneumonia	60 (20)	55 (15)	115 (18)	0.10
- Secondary peritonitis	33 (11)	88 (25)	121 (19)	<0.001
- Urinary tract infection	25 (9)	33 (9)	58 (9)	0.78
- Bloodstream infection	29 (10)	23 (6)	52 (8)	0.11
- Other[c]	59 (20)	81 (23)	140 (22)	0.44
Causative organism				
- Gram-positive bacteria	98 (33)	125 (35)	230 (35)	0.68
- Gram-negative bacteria	106 (36)	161 (45)	260 (40)	0.02
- Fungi and yeasts	23 (8)	13 (4)	25 (4)	0.20
- Viruses	12 (4)	13 (4)	25 (4)	0.84
- Unknown	55 (19)	39 (11)	94 (14)	0.01
Length of stay, days				
- Survivors	9 (4-14)	12 (7-22)	9 (4-18)	<0.001
- Non-survivors	6 (3-12)	5 (3-14.5)	6 (3-14)	0.28
Use of mechanical ventilation	253 (86)	341 (96)	594 (91)	<0.001
Use of renal replacement therapy	37 (13)	131 (37)	168 (26)	<0.001
ICU mortality	57 (19)	148 (42)	205 (32)	<0.001

ICU: intensive care unit. APACHE: Acute Physiology and Chronic Health Evaluation. Continuous variables are expressed as median (IQR). Dichotomous and categorical variables are presented as frequency (%). [a] Cardiovascular disease includes chronic cardiovascular insufficiency, congestive heart failure, myocardial infarction, stroke, cerebrovascular disease and peripheral vascular disease. [b] Immune deficiency includes the presence of acquired immune deficiency syndrome (AIDS) or asplenia, the chronic use of immunosuppressive drugs, exposure to chemo- or radiotherapy in the last 12 months, and any other documented humoral or cellular deficiency. [c] Other infections included for example intra-abdominal infections other than secondary peritonitis, skin or soft tissue infections, infections of the central nervous system, gastro-intestinal infections, upper and lower respiratory tract infections other than community-acquired pneumonia and hospital-acquired pneumonia, and post-operative wound infections.

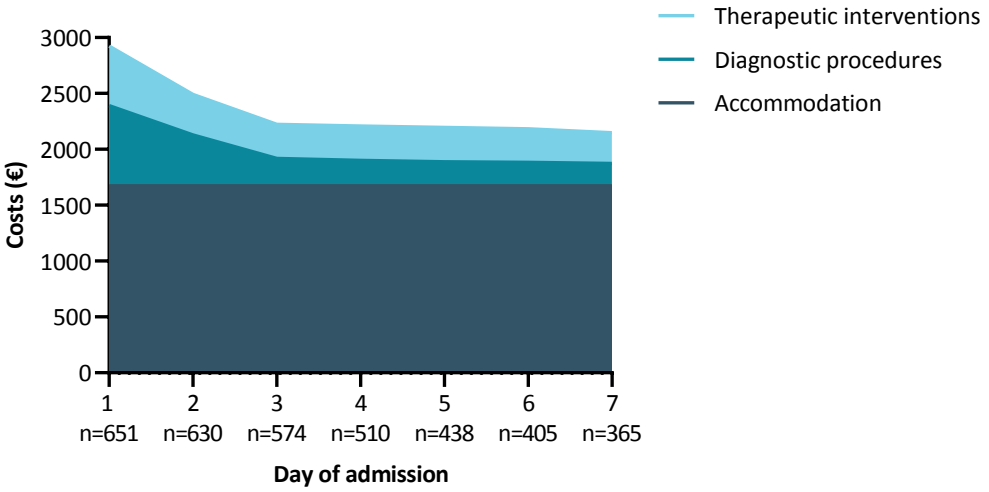
The daily expenditure on both diagnostic procedures and therapeutic interventions was higher during the first two days of admission than on consecutive days (Figure 2). By the end of the second day of ICU admission, the additional costs for diagnostic procedures and therapeutic interventions had accumulated to €732 and €300, respectively.

Table 2. Resource use and costs of treatment for ICU patients with severe sepsis and septic shock

Cost component	Unit of accounting	Price per unit (€)	Usage per patient per day (units of accounting)	Costs per patient per day (€)	Costs per patient per admission (€)
Accommodation (13)	Day		NA		
- Hotel		83		83	664 (332-1411)
- Overhead		572		572	4,576 (2,288-9,724)
- Capital		206		206	1,648 (824-3,502)
- Personnel		777		777	6,216 (3,108-13,209)
- Disposables		36		36	288 (144-612)
Diagnostics: laboratory[a]	Test				
- Blood chemistry		1.5 (1.3-1.7)	35 (26-50)	116 (64-190)	1277 (704-2,439)
- Microbiology		9 (10-11)	0 (0-8)	0 (0-80)	586 (271-1,125)
Diagnostics: radiology[b]	Procedure				
- Standard[c]		44 (44-44)	0 (0-1)	0 (0-44)	117 (88-342)
- Advanced[d]		182 (165-208)	0 (0-0)	0 (0-0)	208 (0-489)
Therapeutics					
- Antibiotics[e]	DDD	7 (2-11)	3 (0-7)	9 (1-32)	84 (31-293)
- Other drugs	Dose[f]	6 (2-19)	0.8 (0.5-1.4)	44 (21-155)	407 (120-1,349)
- Fluids	Litre	56 (29-90)	1 (0.4-1.8)	5 (2-12)	94 (56-170)
- Nutrition	Litre	24 (16-36)	1.4 (1-1.9)	13 (5-17)	77 (16 -219)
- Blood products	Unit	215 (215-557)	0 (0-0)	0 (0-0)	215 (0-1,186)
- Source control[g]	Procedure	691 (471-1,276)	0 (0-0)	0 (0-0)	0 (0-0)[h]

ICU: intensive care unit. NA: not applicable. DDD: defined daily dose. All data are expressed as median (IQR). The IQR for price per unit and costs per patient per day for the component of accommodation is not shown as it is not applicable. [a] The number of specimens could not be separated for blood chemistry and microbiology, therefore the unit of accounting is presented per test. The median number (IQR) of specimens for total laboratory was 3 (2-5). [b] Interventional radiology was included under source control procedures. [c] Including X-rays and ultrasounds. [d] Including all MRI scans, CT scans, PET scans, angiography, pyelography, venography and Duplex ultrasonography. [e] Costs for antibiotic prophylaxis were included under 'other'. [f] Total dose per day per 1,000 units of measurements (including millilitres and milligrams). [g] Including surgical procedures and interventional radiology undertaken to treat (complications of) sepsis in the department of surgery. [h] Median costs per patient per admission were €0 because it was utilized in less than 25% of admissions, but mean costs per patient per admission were €350.

Figure 2. Mean costs per day in the first week of admission



Depicted are the health care costs for patients with severe sepsis and septic shock in the first 7 days of ICU admission.

Patients with septic shock had significantly higher costs per day and per admission compared to patients with severe sepsis (Table 3). The additional costs per day and per admission were €69 (95%CI €37-€100) and €8,355 (95%CI €3,400-€13,367), respectively. This was mainly caused by a longer length of stay of these patients. Differences in costs per admission among patients with different sites of infection were borderline significant, with patients having secondary peritonitis representing the most expensive group. Patients with sepsis caused by fungi or yeasts had twofold higher costs than those with bacterial infections. Furthermore, non-survivors had significantly higher costs per day than survivors, the additional costs were €239 (95%CI €206-€272). But as non-survivors had a shorter length of stay, overall costs per admission did not differ significantly by survival status.

Male gender, younger age, higher APACHE IV score, and the presence of chronic obstructive pulmonary disease and diabetes mellitus were all associated with increased costs in univariable analyses (Table 4). However, none of these variables remained independently associated with costs in multivariable analyses, nor contributed significantly to the total explanatory power of the final cost prediction model. In contrast, the site of infection and its causative organism, the presence of shock at admission, and prior immune deficiency remained independently associated with increased expenditure in the multivariable analysis. Overall, using information available in the first 24 hours of ICU admission and an optimized multivariable prediction model, we could explain only 11.2% of the observed variability in health care expenditure between patients.

Table 3. Costs per day and per admission for subgroups of patients

	N	Costs per day (€)	P-value	Costs per admission (€)	P-value
Total	651	2,250 (2,235-2,266)	NA	29,102 (26,598-31,690)	NA
Disease severity			0.04		<0.001
- Severe sepsis	294	2,208 (2,188-2,227)		24,520 (21,288-27,973)	
- Septic shock	357	2,277 (2,255-2,298)		32,875 (29,615-36,281)	
Site of infection			<0.001		0.05
- Community-acquired pneumonia	165	2,152 (2,129-2,175)		27,913 (23,467-33,105)	
- Hospital-acquired pneumonia	115	2,266 (2,239-2,294)		33,162 (26,456-40,721)	
- Secondary peritonitis	121	2,270 (2,236-2,306)		35,554 (29,239-42,352)	
- Urinary tract infection	58	2,153 (2,109-2,198)		18,331 (14,509-22,360)	
- Bloodstream infection	52	2,260 (2,213-2,313)		30,471 (22,715-39,535)	
- Other[a]	140	2,375 (2,327-2,427)		25,546 (22,121-29,168)	
Causative organism			<0.001		<0.001
- Gram-positive bacteria	223	2,217 (2,194-2,241		28,910 (24,787-33,339)	
- Gram-negative bacteria	267	2,206 (2,182-2,231)		27,014 (23,820-30,261)	
- Fungi and yeasts	42	2,440 (2,396-2,241)		59,600 (43,216-78,511)	
- Viruses	25	2,472 (2,389-2,560)		38,755 (28,368-51,186)	
- Unknown	94	2,201 (2,161-2,242)		19,294 (15,783-23,209)	
Survival status			<0.001		0.08
- Survivors	446	2,179 (2,163-2,195)		28,824 (26,141-31,522)	
- Non-survivors	205	2,418 (2,385-2,451)		29,707 (24,854-35,109)	

NA: not applicable. Costs are expressed as mean (bootstrapped 95%CI). [a] Other infections included for example intra-abdominal infections other than secondary peritonitis, skin or soft tissue infections, infections of the central nervous system, gastro-intestinal infections, upper and lower respiratory tract infections other than community-acquired pneumonia and hospital-acquired pneumonia, and post-operative wound infections.

Discussion

In this study, the direct health care expenditure associated with treatment of severe sepsis and septic shock in the ICU was estimated to be almost €30,000 per admission. Variable costs for diagnostic procedures and therapeutic interventions represented only 26% of the total expenditure, whereas the largest component consisted of fixed costs for accommodation, personnel and disposables. Furthermore, we found that patient and illness characteristics at baseline explained only a small part of the observed variability between individuals. Based on these results, it seems impossible to take costs of treatment into account when clinical decisions are made regarding the initiation of ICU treatment. Furthermore, since most independent risk factors for high expenditure are related to fixed patient characteristics, our study does not provide direct approaches to reduce costs. However, improved cost-effectiveness may alternatively be sought in reducing the use of inefficient diagnostic or inefficacious therapeutic procedures, for example by implementing new molecular methods for risk stratification of patients, rapid pathogen detection, and so forth.

Table 4. Multivariable model to explain variability in costs per admission between patients

Independent Variable	Coefficient (€) (95% CI)	R^2 change (%)
Site of infection		6.6
- Urinary tract infection	(reference)	
- Community-acquired pneumonia	8,058 (1,666-14,790)	
- Hospital-acquired pneumonia	11,520 (4,477-19,017)	
- Secondary peritonitis	14,532 (7,159-22,514)	
- Bloodstream infection	11,462 (2,846-21,877)	
- Other[a]	6,048 (206-12,261)	
Causative organisms		2.1
- Unknown	(reference)	
- Gram-positive bacteria	8,960 (2,476-15,368)	
- Gram-negative bacteria	6,731 (1,060-12,217)	
- Fungi and yeasts	36,286 (20,119-54,769)	
- Viruses	19,846 (9,271-31,675)	
Septic shock at admission	8,047 (3,374-12,764)	1.6
Immune deficiency[b]	7,027 (1,309-12,711)	0.8

CI: confidence interval. R^2: explained variance. As an example of interpretation, a patient with secondary peritonitis will be predicted to invoke additional costs of almost €15,000 compared to a patient with a urinary tract infection. [a] Other infections included for example intra-abdominal infections other than secondary peritonitis, skin or soft tissue infections, infections of the central nervous system, gastro-intestinal infections, upper and lower respiratory tract infections other than community-acquired pneumonia and hospital-acquired pneumonia, and post-operative wound infections. [b] Immune deficiency includes the presence of acquired immune deficiency syndrome (AIDS) or asplenia, the chronic use of immunosuppressive drugs, exposure to chemo- or radiotherapy in the last 12 months, and any other documented humoral or cellular deficiency.

Costs of sepsis are of high societal impact. In European countries, approximately 28% of ICU patients have community or hospital-acquired infections (15). Therefore, a major part of the European budget for ICUs is spent on sepsis care. In the Netherlands, for example, an estimated 8,500 patients present to ICUs with severe sepsis or septic shock each year (16). Extrapolating the costs per sepsis case as estimated in the present study, total direct health care expenditure for Dutch society will likely exceed 250 million euros annually. Furthermore, indirect medical costs associated with the sepsis episode due to long-term negative health consequences following ICU treatment, may substantially contribute to the total economic burden of disease from the societal perspective.

Our total cost estimate is higher than reported in two previous European studies that have valued the total costs of sepsis treatment in the ICU at €22,800 and €23,297, respectively (5, 6). As one study exclusively enrolled patients having a length of stay longer than 48 hours, the total expenditure per admission they reported may even have been an overestimation of costs for all-comers with sepsis (5). It seems plausible that expenses for sepsis treatment have risen over the last decade (1, 8), given the expanded possibilities of care due to technological advancements as well as the generally increased

burden of disease due to demographic change. However, after correction for inflation and price level variations between countries, our cost estimate remains only slightly higher than these previous projections. Overall, our results thus seem comparable to earlier findings, indicating that our study sample was representative of sepsis patients treated in other European settings.

In contrast to most previous publications (5, 6), but in agreement with at least one other study (9), we did not find relevant differences in costs for treatment of sepsis in the ICU between survivors and non-survivors. However, we did find higher costs per day for non-survivors as compared to survivors, which probably reflects both a greater severity of illness in these patients and the aggressive attempts of clinicians to prevent death (6). We also did not find that older patients generate lower costs, as was previously reported by others (5, 11). The suggestion by some authors that elderly patients are treated less aggressively in the ICU was thus not confirmed (5).

We found that patient and illness characteristics measured in the first 24 hours in the ICU could not be used to reliably predict which individuals would generate high costs. In fact, four variables remained independently associated with increased costs in our multivariable model but together they could explain only 11% of the observed variability in expenditure between patients. These variables were: the presence of shock at admission, infection caused by fungi or yeasts (and to a lesser extent also viral reactivation of cytomegalovirus and herpes simplex virus), site of infection, and immunodeficiency. Urinary tract infections generated the lowest costs and secondary peritonitis the highest costs, followed by hospital-acquired pneumonia and bloodstream infections. This finding matches results of two French studies (5, 9). The significantly higher costs that we found for immune deficient patients were mainly caused by higher costs for therapeutic interventions, which reflects the difficulties in treating infection in these patients. Other variables that we a-priori selected based on literature, did, in contrast to earlier studies (5, 11), not remain independently associated with costs in our model. These variables were patient characteristics such as age and the presence of comorbidities, but also surgical admission was not associated with costs, although this was suggested in three earlier studies (5, 6, 11).

Strengths of this study relate to the use of data collection as part of a multicentre, prospective cohort study and the accuracy that this entails. This enabled us to provide detailed information about individual cost components, to compare costs between subgroups, and to investigate variability in costs between individuals. However, a limitation of this study is that we derived the daily costs for accommodation, personnel and disposables from literature. As a result, the intensity of care of individual patients was not reflected in the health care expenditure. However, a stable patient does acquire less attention from nurses and physicians, but this does not entail lower costs because personnel is present anyway. Secondly, both participating hospitals use protocols for selective decontamination of the digestive tract in ICU patients.

Although this intervention has been shown to reduce the incidence of ventilator-associated pneumonia and mortality, use of antibiotic prophylaxis generates costs per se (17). Nonetheless, selective decontamination of the digestive tract was shown to be cost-effective (18). Therefore, the adherence to these protocols may hamper the generalizability of our findings.

Conclusions

In conclusion, total health care costs per ICU admission for severe sepsis or septic shock are almost €30,000 per patient. Costs were poorly predictable by patient and illness characteristics measured at baseline, which limits the development of potential costs saving strategies based on patient profiling at ICU admission.

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4

Chapter 4

Chronic healthcare expenditure in survivors of sepsis in the intensive care unit

Maria E. Koster-Brouwer
Kirsten van de Groep
Wietze Pasma
Hugo M. Smeets
Arjen J.C. Slooter
Dylan W. de Lange
Diederik van Dijk
Tom van der Poll
Marc J.M. Bonten
Olaf L. Cremer
On behalf of the MARS consortium

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Abstract

Purpose Direct expenses associated with an intensive care unit (ICU) admission for sepsis equal approximately €30,000, but total health care costs are likely to be much higher because survivors frequently suffer from long-term sequelae. We estimated expenditure before and after sepsis, and assessed its correlation with health-related quality of life (HRQoL).

Methods We analyzed one-year survivors of sepsis enrolled in a prospective cohort study in two tertiary ICUs in the Netherlands in 2011 and 2012. Health care utilization and costs were derived from a Dutch insurance company database. Trends in expenditure were assessed using interrupted time series analysis. HRQoL was measured using self-reported EQ-5D in a subgroup of patients.

Results We analyzed 357 (96%) of 373 eligible patients. Comparing two-year periods before and after sepsis, we observed significant increased utilization of hospital care, long-term (home) care, allied health care, and mental care. Mean total costs per month were €1,035 (SD 2,009) and €3,533 (SD 5,190) before and after sepsis, respectively. After adjustment for baseline trends, increase in monthly expenditure following sepsis was estimated at €2,281 (95%CI 1,755 to 2,807, p=0.005). During follow-up, regression towards baseline reimbursement levels was observed in all subgroups, except in elderly with comorbidities. However, overall monthly expenditure two years after the event was still €1,690 (95%CI 601 to 2,779) higher than predicted by baseline trends. Mean HRQoL was 0.70 (SD 0.26), with higher expenditure being negatively correlated with favorable outcome (Spearman's correlation coefficient -0.439, p<0.001).

Conclusions Sepsis survivors generate substantial costs up to at least two years after the event which cannot be explained by pre-existing trends.

Introduction

Although sepsis represents a severe but transient disease process, survivors of sepsis-related critical illness are frequently confronted with a series of long-term sequelae (1). These may entail physical limitations, cognitive impairments, and various psychiatric symptoms, including post-traumatic stress disorder and depression (2, 3). Together, these conditions have been coined as the post intensive care syndrome (PICS) (4). Although the impact of PICS on health-related quality of life (HRQoL) is reported to be most pronounced in patients already having pre-existing comorbidities (5), also previously healthy patients may be severely impacted (2). As the acute mortality of sepsis has been gradually declining over the past decades (6, 7), it is likely that increased numbers of survivors will be confronted with these negative long-term health consequences in the future.

The direct costs associated with an intensive care unit (ICU) admission for severe sepsis or septic shock are highly variable, but average approximately €30,000 per case (8, 9). However, the true economic burden of sepsis-related critical illness to society is likely to extend far beyond the acute phase of the disease, because survival with PICS and an increased vulnerability to develop for example new infections may both incur a need for long-term care and hospital (re)admissions (10). Indeed, previous studies have reported increased health care utilization (11, 12) and high costs (approximately $34,000, mainly due to rehospitalizations) in the first year after sepsis (13). However, these studies did not account for the premorbid status of patients, precluding accurate estimations of the change in health status and expenditure attributable to PICS. Furthermore, within the limits imposed by a one-year observation window it is not possible to assess the rates at which excess costs incurred immediately after the sepsis event may eventually return to baseline levels over time. Finally, it seems plausible that advanced age and the presence of pre-existing comorbidities would significantly impact ongoing resource use after discharge, but this association has also not been formally studied.

To estimate costs associated with PICS in survivors of critical illness we measured health care utilization during a two-year period following an ICU admission for sepsis, while correcting for trends in expenditure already present during the two years before the event. In addition, we explored the impact of age and comorbidities on changes in health care expenditure. Finally, in a subgroup of patients we investigated the association between health care expenditure and long-term HRQoL.

Methods

Patients

Subjects were enrolled as part of the Molecular Diagnosis and Risk Stratification of Sepsis (MARS) cohort, a prospective study in the mixed medical-surgical ICUs of the Academic Medical Center Amsterdam and University Medical Center Utrecht (UMCU) in the Netherlands (14). For the current analysis we included consecutive patients admitted to the ICU because of sepsis in 2011 or 2012, who had survived for more than a year following ICU admission. Sepsis was defined according to SCCM/ACCP guidelines (15). Overall, 99% of included patients had a sequential organ failure assessment (SOFA) score ≥2 at ICU admission, which approximates the recently updated consensus definitions for sepsis (16). Sepsis onset had to have occurred within a 48-hour time window surrounding ICU admission. The post-hoc likelihood of true infection had to be rated at least possible according to retrospective physician assessment based on validated definitions (14). Survival status was queried from the Dutch municipal population registers 12 to 14 months after discharge. Ethical approval for the study was provided by the Medical Ethics Committees of participating centers, including a waiver for informed consent (IRB numbers 14-095/C, 10-056, 10-006).

Data collection

Clinical information collected during the MARS study was combined with cost data from a Dutch insurance company, the Achmea Health Database. This database contains information on reimbursed health care of 4.3 million Dutch citizens over prolonged periods of time, and represents an unbiased population sample in urbanized areas of the Netherlands (17). To ensure the privacy of included patients, records were pseudonymized by a trusted third party (ZorgTTP, Houten, the Netherlands) before data were combined. Information from the two-year period immediately before the sepsis episode and the two-year period afterwards was queried, totaling 4 years of reimbursement data. We categorized expenditure into costs for hospital care, long-term (home) care (including rehabilitation facilities, nursing homes, and home care), pharmaceutical care, consultations of the general practitioner, allied health care (including physical therapy, occupational therapy, speech therapy, and dietetics), and mental health care (including consultations of social workers, psychologists, and psychiatrists).

Subsequently, in one of the two participating centers (UMCU) only, patients were asked to complete a written follow-up questionnaire to assess HRQoL. Participants or their relatives were requested to rate the degree of disability for each dimension of the EuroQol five dimensions (EQ-5D-3L) questionnaire, i.e. mobility, self-care, daily activities, pain and discomfort, and anxiety and depression. The results of the EQ-5D are translated into a health status index ranging from 0 to 1, in which 0 stands for death and 1 for perfect health (18).

Statistical analyses

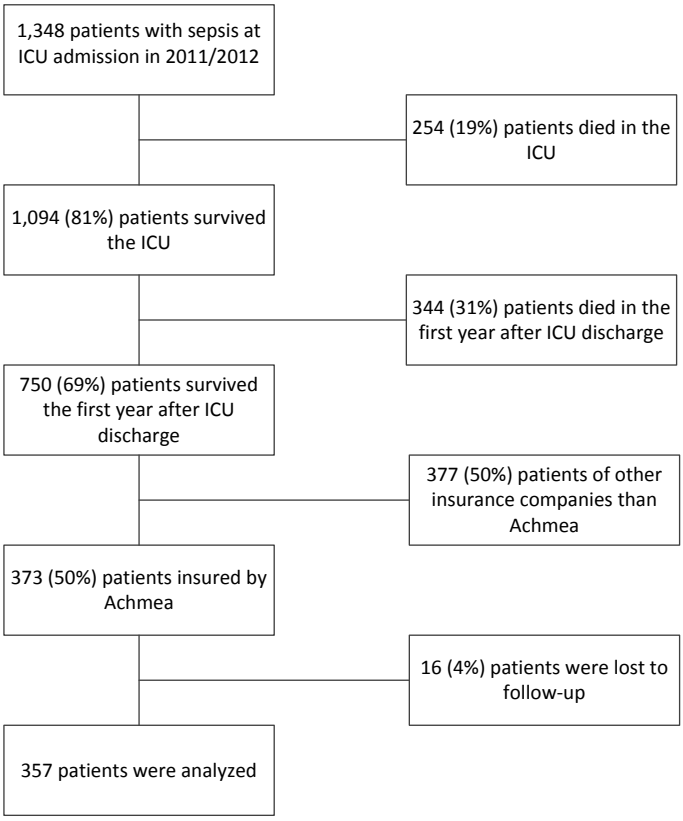
Among the data set totaling four years of reimbursement information, we excluded all costs incurred during the six months directly preceding and following the sepsis episode in order to eliminate potential carry-over effects from charges that were directly related to the sepsis event rather than to chronic health service utilization. The proportions of patients using certain types of care before and after the sepsis event were first compared using McNemar's test. We then used interrupted time series (ITS) analysis to compare reimbursed costs incurred during the -24 to -6 month period before sepsis with the +6 to +24 month period after the event. This method uses segmented linear regression analysis to assess whether there is a change in the population average of health care expenditure in the period before compared to the period after the sepsis episode, while correcting for autocorrelation between consecutive reimbursement data. ITS analyses estimates both level and trend effects. The baseline trend represents the underlying change in monthly health care expenditure in the population. Trend effects thus reflect a change in slope after the event compared to this baseline trend (19, 20). Level effects represent the abrupt change in monthly costs due to the event, in addition to the estimated expenditure based on continuation of the baseline trend until the time point of interest (month +6). Furthermore, at the end of follow-up (month +24) we compared the expected population average of health care expenditure according to observed baseline trends with actual expenses, using methods as described previously (20). ITS analyses were performed for the total cohort, as well as for subgroups of patients. Based on results from studies which investigated long-term consequences of sepsis and/or ICU stay (2, 5, 10, 13), we divided the cohort into four strata: patients aged younger and older than 65 with and without comorbidities. The presence of comorbidities was defined as a Charlson comorbidity index greater than 0.

In addition to the ITS analysis, we analyzed the extent to which health care expenditure was correlated with HRQoL using Spearman's rank correlation coefficient in patients for whom follow-up was available. Cost data are presented as means with standard deviations, as this enables readers to extract absolute values for total health care expenditure at a group level. All other data are presented as medians (IQR) or absolute numbers (%) as appropriate. All analyses were performed using SAS Enterprise Guide 5.1 (SAS Institute, Cary, NC), p-values <0.05 were considered statistically significant.

Results

Among 1,348 unique sepsis admissions, 750 (69%) patients survived beyond one year; of these, 373 (50%) remained after exclusion of individuals who were not insured by Achmea (Figure 1).

Figure 1. Flowchart of patient inclusion



ICU: intensive care unit.

Patients carrying health insurance from Achmea did not differ from remaining patients (who were insured by other companies) when comparing baseline characteristics (data not shown). Insufficient data were available to assess health care expenditure in 16 patients and these were therefore considered lost-to-follow-up. Baseline characteristics of the 357 included patients are shown in Table 1.

Health care expenditure

Figure 2 displays the population average of monthly health care expenditure in the two-year periods immediately preceding and following a sepsis episode in the ICU.

Unadjusted mean monthly health care expenditure was €1,035 (SD 2,009) and €3,533 (SD 5,190) before and after the sepsis event, respectively (crude cost difference €2,498, SD 4,678, $p<0.001$). This increase resulted predominantly from greater expenses for hospital care, long-term (home) care, and mental health care (Table 2).

Table 1. Patient and disease characteristics of 1-year sepsis survivors

	Total cohort (n=357)
Gender, male	210 (59)
Age, years	61 (49-70)
Charlson comorbidity index	3 (0-8)
Surgery prior to ICU admission	109 (31)
Previous ICU admission	48 (14)
Nosocomial origin of infection	135 (38)
Septic shock at admission	76 (21)
Site of infection	
- Community-acquired pneumonia	103 (29)
- Hospital-acquired pneumonia	83 (23)
- Abdominal sepsis	52 (15)
- Blood stream infection	23 (6)
- Urinary tract infection	21 (6)
- Othera	75 (21)
APACHE IV score	70 (54-91)
Worst SOFA score observed in the ICU	8 (5-10)[b]
Mechanical ventilation	299 (84)
Renal replacement therapy	46 (13)
Length of ICU stay, days	4 (2-9)
Length of hospital stay, days	22 (12-42)

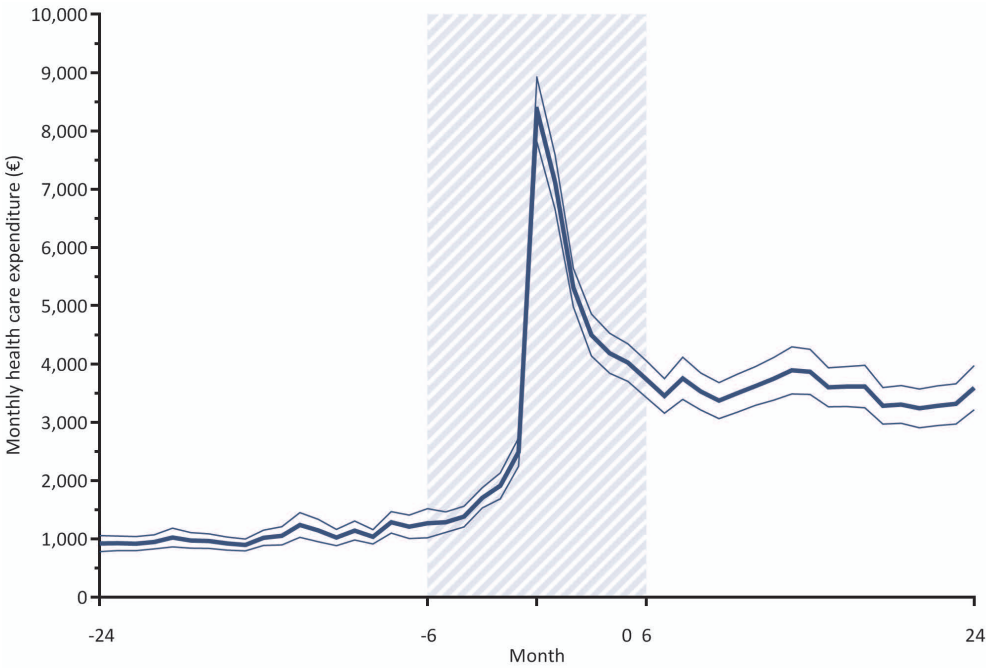
Dichotomous variables are presented as frequency (%), continuous variables are presented as median (IQR). ICU: intensive care unit, APACHE: Acute Physiology and Chronic Health Evaluation, SOFA: Sequential Organ Failure Assessment. [a] Including, but not limited to, wound or skin infections (n=18), infections of the central nervous system (n=11), upper respiratory tract infections (n=9), and ventilator-associated pneumonia (n=7). [b] 25 missing values.

Table 2. Monthly resource use and health care expenditure preceding and following a sepsis episode (n=357)

	Resource use			Expenditure (€)a	
	Before sepsis	After sepsis	P-value	Before sepsis	After sepsis
Hospital care	15 (4)	19 (5)	<0.001	781 (1,831)	2,382 (4,693)
Long-term (home) careb	1 (0)	8 (2)	<0.001	13 (80)	821 (1,852)
Pharmaceutical care	18 (5)	18 (5)	0.527	193 (451)	231 (457)
GP consultations	18 (5)	18 (5)	0.873	8 (8)	8 (8)
Allied health carec	6 (2)	9 (3)	<0.001	27 (87)	40 (88)
Mental health cared	1 (0)	3 (1)	<0.001	14 (131)	52 (344)

Resource use is presented as the number of patients (%) utilizing the listed type of care at least once during the two-year periods before and after sepsis, respectively. Health care expenditure is presented as the mean (SD) of reimbursed costs within the listed category of care. GP: general practitioner. [a] P-values are not shown as these are crude comparisons, without adjustment for baseline trends. [b] Including rehabilitation facilities, nursing homes, and home care. [c] Including physical therapy, occupational therapy, speech therapy, and dietetics. [d] Including consultations of social workers, psychologists, and psychiatrists.

Figure 2. Monthly health care expenditure in the 2 years preceding and following a sepsis episode (n=357)



Monthly health care expenditure is depicted as mean (SE). The shaded area represents the 6-month periods leading up to and immediately trailing the sepsis event; costs incurred during this time interval were excluded from further analysis.

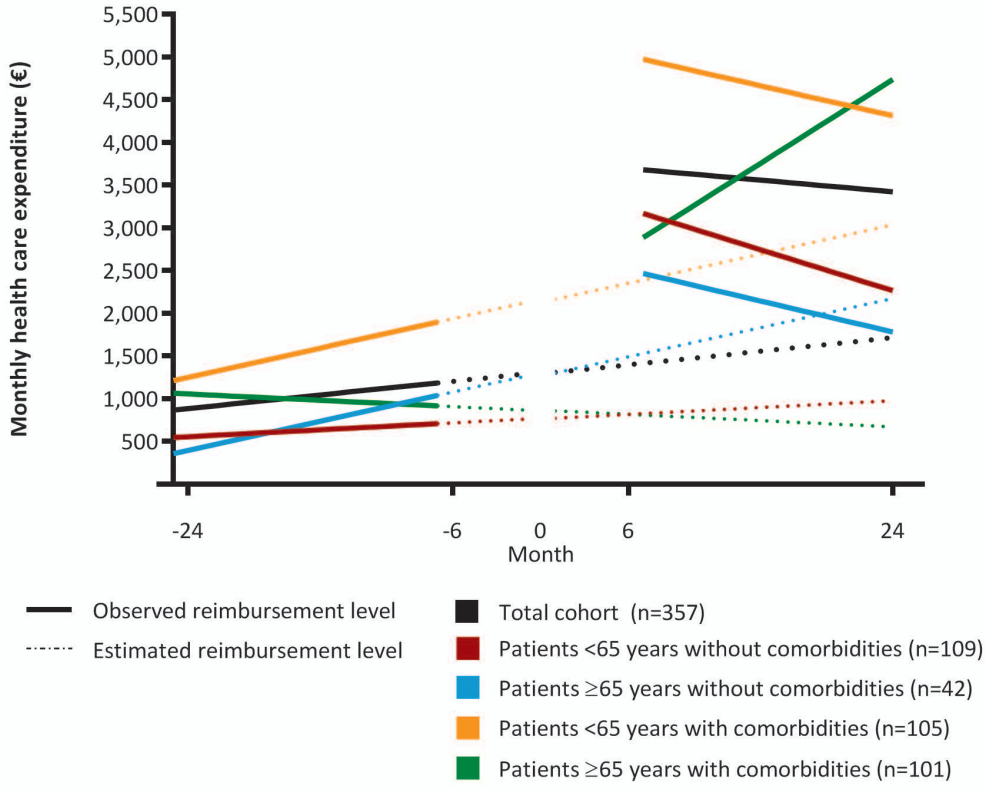
Results of the ITS analyses are presented in Table 3.

Table 3. Estimations of step increases and trend effects following a sepsis episode (n=357)

	Monthly health care expenditure (€)	95% CI	P-value
Intercept	867	628 – 1,107	<0.001
Baseline trend	18	-5 – 40	0.115
Level effect	2,281	1,755 – 2,807	<0.001
Trend effect	-33	-64 – -2	0.040

At the start of the baseline period the average monthly reimbursement level in the cohort was estimated to be €876 (95%CI 628 to 1,107, P<0.001), with a non-significant slope indicating a rise in expenditure by €18 (95%CI -5 to 40, p=0.115) each month. After correction for this baseline trend, the observed increase in monthly health care expenditure following sepsis remained (adjusted cost difference €2,281 (95%CI 1,755 to

Figure 3. Monthly health care expenditure before and after a sepsis episode in subgroups of patients (n=357)



2,807, p<0.001). A calculation with correction for observed temporal trends before and after the sepsis event showed that monthly expenditure was still increased by €1,690 (95%CI 601 to 2,799) two years after the sepsis event.

Subgroup analyses

The results of the ITS analyses per subgroup based on age and comorbidities are plotted in Figure 3.

Results of ITS analyses of differences between subgroups are available in the supplementary material (Appendix I, Table S1). Regardless of age, patients with comorbidities had higher monthly reimbursement levels at baseline than patients without comorbidities, but there were no significant differences in baseline trend between any of the subgroups. Directly after the sepsis event, older patients incurred lower reimbursement levels than younger patients regardless of their comorbid status. Decreasing trends in monthly health care expenditure after sepsis were observed for 3 subgroups, possibly indicating that these patients were still recovering from their

acute illness up to at least two years after the event. The opposite was seen in elderly patients with comorbidities in whom costs continued to rapidly increase, which might indicate an escalating effect in these frail elderly.

Health-related quality of life

Among the total cohort of sepsis survivors, 140 (39%) patients received a follow-up questionnaire and HRQoL could be calculated for 90 (64%) of them. Patients who returned a questionnaire differed from those who were either not approached at all or did not respond to the EQ-5D query by having higher age, more comorbidities at baseline, and a greater severity of illness at ICU admission (data not shown). Approximately 14 months after the sepsis episode (median follow-up 417 days, range 302–554) the mean health status index of the surveyed patients was 0.70 (SD 0.26). This is lower than the mean health status index for Dutch citizens of comparable age, which is 0.85 (SD 0.18) (21).

Although costs were highly variable across the entire range of HRQoL measurements, formal analysis elicited significant negative correlation (Spearman's correlation coefficient -0.439, $p < 0.001$), indicating that higher health care expenditure was associated with worse HRQoL (Appendix II, Figure S1). Furthermore, greater step increases in expenditure following the sepsis episode (rather than absolute costs) were also significantly correlated with lower HRQoL (Spearman's correlation coefficient -0.410, $p<0.001$; data not shown).

Discussion

Patients who survived sepsis in the ICU demonstrated an increase in long-term health care expenditure by almost €2,300 per month when we compared their reimbursement rates after sepsis with predicted expenditure according to baseline trends. These expenses slowly decreased during subsequent follow-up in most individuals, indicating that patients were still recovering. However, even two years after the sepsis episode monthly health care expenditure was still increased by €1,690 overall. Furthermore, in elderly survivors with pre-existing comorbidities costs continued to increase as time progressed. As sepsis itself is an ephemeral illness, it is plausible that these excess costs are attributable to PICS, rather than to prior disease processes which may have triggered infection onset.

In recent years, awareness of PICS has grown significantly (4). As both the burden of disease for individual patients and the financial burden for society seems substantial, there is a clear need for strategies that may prevent the development of these late sequelae of critical illness. Because costs increased across all categories of care in our study, we suggest that such interventions should have a broad and multidisciplinary approach.

The results of our study are in line with studies that have previously reported the development of new impairments (2), increased health care utilization (11, 12), and high ongoing health care costs (13, 22) after treatment of sepsis in the ICU, as well as in a general critical care populations. However, using highly detailed reimbursement data covering extended periods of time both before and after the sepsis episode, we were not only able to better quantify the economic burden of disease, but could also precisely describe the impact of a sepsis episode on long-term utilization of hospital, long-term (home), mental and allied health care. We did find high impact of ICU treatment for sepsis on expenditure in all strata of age and comorbidity. Furthermore, pre-existing comorbidities showed to be an essential feature, since young patients with comorbidities had the highest increase after sepsis and elderly with comorbidities had the most pronounced change in slope during follow-up. This is consistent with previous indications that the impact of ICU treatment is greater in patients with pre-existing suboptimal health (5). However, a study which assessed physical functioning before and after sepsis did not find these higher impact in patients with baseline limitations, but the researchers already posed that this might be a ceiling effect of the used measurement instrument (2). Overall, we found approximately threefold higher estimates of hospital health care expenditure than previously estimated (22). Aside from the fact that we included also outpatient care in this cost category, an additional explanation might be that we exclusively included patients with sepsis who had probably a higher severity of disease and a longer ICU and hospital length of stay when compared to the general ICU population studied by Lone et al. (22). Therefore, it is plausible that the impact of PICS in our cohort is more significant, which would explain the higher financial burden after ICU discharge that we observed.

To our knowledge, this is the first study comparing detailed information about chronic health care expenditure before and after sepsis using the patient as its own control. This design enabled us to investigate the impact of the sepsis episode on trends in health care expenses already present, rather than merely providing a point-estimate of long-term expenditure. Since insurance companies can provide an accurate overview of all reimbursed chronic health resource utilization, and since higher expenditure is negatively correlated with HRQoL, cost data will accurately reflect the long-term impact of critical illness. Although the exact expenditures might not be directly generalizable to countries with other health care systems, the patterns in costs will likely be very similar between patients in different settings.

As a consequence of the study design, some considerations have to be taken into account when interpreting the results of our study. Firstly, we aimed to quantify costs associated with PICS. For this purpose we deliberately focused on one-year survivors of sepsis. However, some patients who died within a year after they had initially been discharged alive from the hospital may have generated high costs in the period immediately before death which were not accounted for in our study (23). Due to this,

and to the fact that we also did not include costs for productivity losses, the total economic impact of sepsis in the ICU from a societal perspective will be higher. Secondly, 42 patients (12% of the total study cohort) had some period without any registered care by the end of follow-up. This could either be due to death in the second year after discharge, or to complete non-use of health care services during this period. In either case, no charges were incurred and these data points were therefore analyzed as such. This is a conservative approach, as it will always result in lower cost estimations when compared to other scenarios, for example handling missing values by replacing them with the mean health care expenditure of the cohort for that month. Furthermore, cost data are typically skewed to the right, implying that relatively few patients with excessive expenses will drive the mean. Nonetheless, we used ITS analyses based on means instead of alternative approaches (e.g., based on medians) because mean values enable better translation of study results to reimbursement rates on a population level, and are thus more informative for health policy decision making.

In conclusion, the economic burden of sepsis reaches far beyond initial ICU treatment. Even after two years, survivors of sepsis continue to generate substantial health care expenditures. Furthermore, high resource utilization after critical illness seems to be a representation of a chronically diminished health status.

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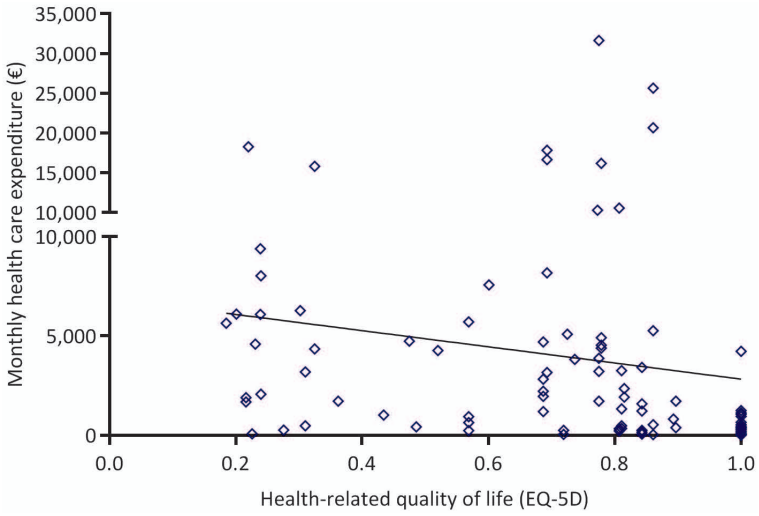
SUPPLEMENTARY MATERIAL

Appendix I: Table S1. Results of interrupted time series analyses for subgroups of sepsis patients (n=357)

	Estimate	95% CI	P-value
Patients ≥65 years without comorbidities (n=42)			
Intercept	-181	-519 – 157	0.286
Baseline trend	31	-1 – 62	0.054
Level effect	-1,453	-2,195 – -711	<0.001
Trend effect	-19	-63 – 25	0.392
Patients <65 years with comorbidities (n=105)			
Intercept	709	331 – 1,088	<0.001
Baseline trend	21	-14 – 56	0.229
Level effect	743	-87 – 1,574	0.078
Trend effect	-41	-90 – 8	0.101
Patients ≥ with comorbidities (n=101)			
Intercept	422	123 – 721	0.007
Baseline trend	-3	-30 – 25	0.854
Level effect	-696	-1,352 – -40	0.038
Trend effect	155	116 – 194	<0.001

Cost estimates in this table reflect differences between each listed subgroup and a reference category consisting of patients younger than 65 years without comorbidities (n=109). Absolute costs per subgroup are shown in figure 3. Cost estimates are expressed as monthly health care expenditure in euros.

Appendix 2: Figure S1. Correlation between mean monthly health care expenditure and self-reported health-related quality of life one year after a sepsis episode (n=90)




Lieve tante Marjan,
Vanharte beterschap
hoop dat u snel
weer opknapt
liefs Johan. Heleen
en de kids

Part III

Modelling long-term sequelae

5

Chapter 5

The association of hyperinflammation and tissue hypoxia with new-onset type 2 diabetes mellitus in survivors of critical illness

Maria E. Koster-Brouwer
David S.Y. Ong
Meri R.J. Varkila
Martinus P. van Ginkel
Arjen J.C. Slooter
Dylan W. de Lange
Diederik van Dijk
Eelco J.P. de Koning
Marc J.M. Bonten
Olaf L. Cremer

In submission

Abstract

Objective Patients in the intensive care unit (ICU) often have dysregulated glucose metabolism, which has been linked to exposure to hyperinflammation and tissue hypoxia during critical illness. Furthermore, ICU survivors are prone to developing type 2 diabetes mellitus (DM2). We investigated whether these associations are etiologically related.

Design Nested case-control study.

Setting A tertiary ICU in the Netherlands.

Patients We studied one-year ICU survivors without prior diabetes who had been admitted to the ICU between 2009 and 2016. Patients were followed-up using a written questionnaire, and new-onset DM2 was identified based on self-reported medication use. Cases were matched to controls based on age, sex, body-mass index, and transplant status. We used multivariable logistic regression to model the association of hyperinflammation (i.e., days with CRP >100 mg/L) and tissue hypoxia (i.e., days with lactate >2 mmol/L) during ICU stay with DM2 incidence.

Interventions None

Measurements and main results The incidence proportion of new-onset DM2 in our source population (n=3185) was 2.4% (95%CI 2.0-3.0%) during a follow-up period of 426 (IQR 405-446) days. Among 78 cases, 76 subjects could be matched to 152 controls. Cases more frequently had increased CRP levels during the first 3 days in ICU (p=0.048), but not more lactatemia (p=0.523). In multivariable analysis, hyperinflammation remained independently associated with new-onset DM2 (OR 1.98, 95%CI 1.10-3.55; p=0.022), but not tissue hypoxia (OR 1.02, 95%CI 0.57-1.84; p=0.936).

Conclusions Severe inflammation, not tissue hypoxia, during critical illness might increase the risk of new-onset type 2 diabetes after ICU discharge.

Introduction

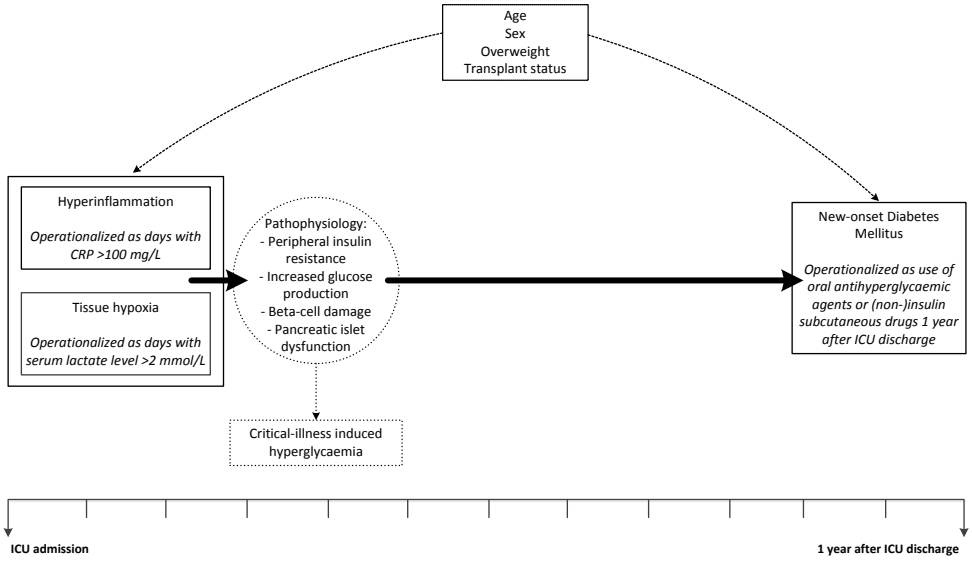
Survivors of critical illness are often confronted with impaired health in the period after intensive care unit (ICU) discharge. Whilst recovered from their acute disease, patients may suffer from cognitive impairment, physical immobility, and mental dysfunction (1-3). Furthermore, ICU survivors are susceptible to onset or deterioration of chronic comorbidities, including development of new cardiovascular events, worsening of renal injury, and exacerbation of chronic obstructive pulmonary disease (4-7).

Against this background, several studies have also reported higher incidences of new-onset type 2 diabetes mellitus (DM2) in ICU survivors than may be expected in the general population (8-11). This holds particularly true in subjects who did experience a period of critical illness-induced hyperglycaemia (12-16). However, any possible etiological link between these observations is prone to be confounded by lifestyle factors and the presence of (covert) underlying disease processes that led to the ICU admission in the first place. Although raised blood sugar in and of itself is likely to be merely a prognostic factor (rather than an etiologic cause) for new-onset DM2, it might be that some of the pathophysiological mechanisms underlying critical illness-induced hyperglycaemia also play a role in the pathogenesis of DM2 in ICU survivors.

During critical illness, hepatic gluconeogenesis increases in response to stress, causing higher blood glucose levels than in health (17-19). In patients with severe inflammation this process is even more pronounced (18, 19). Due to the release of tumor necrosis factor (TNF) α, interleukin (IL) 1, the adipocyte-specific factor resistin, and several other pro-inflammatory mediators, insulin fails to effectively inhibit gluconeogenesis and glycogenolysis, and insulin-stimulated glucose uptake becomes impaired (19). Simultaneously, oxidative stress and tissue hypoxia (such as commonly induced by circulatory shock) can also lead to hyperglycaemia (19, 20). The latter is likely due to the production of nitric oxide and other reactive oxygen species, which trigger an unfolded protein response causing endoplasmic reticulum stress-induced beta-cell death in the pancreas (20, 21). In addition to these inflammatory and hypoxia-mediated mechanisms, hyperglycaemia may be further aggravated by several iatrogenic causes, such as the administration of carbohydrate-rich nutrition, or the concurrent use of corticosteroids (17, 19). These processes and their possible associations with the development of DM2 in ICU survivors are depicted in Figure 1.

In the present study, we aimed to determine the incidence proportion of new-onset DM2 after critical illness and identify risk factors for its development. Specifically, we hypothesized that hyperinflammation and tissue hypoxia are important etiologic factors underpinning both the temporary dysregulation of glucose metabolism during critical illness itself, as well as ongoing peripheral insulin resistance and pancreatic islet dysfunction that causes new-onset DM2 in ICU survivors.

Figure 1. Schematic overview of the modelled association between disease-mediated factors hyperinflammation and tissue hypoxia and new-onset type 2 Diabetes Mellitus



Schematic overview of the modelled association between hyperinflammation and tissue hypoxia during critical illness and the occurence of Diabetes Mellitus after ICU discharge. The rectangles on both sides represent independent and dependent variables that were modelled. The upper rectangle includes variables that are supposed to be confounders or effect modifiers of the association.The circle represents an unobserved variable which is hypothesized to be the pathophysiological mechanism leading to critical-illness induced hyperglycaemia as well as new-onset diabetes mellitus.

Materials and methods

Study population

We studied all patients who had been admitted to the ICU of the University Medical Center Utrecht between June 2009 and July 2016 and survived the first year after ICU discharge, with the exception of short-stay patients admitted after uncomplicated elective cardiothoracic surgery. We excluded patients who were known to have prior DM or had been newly diagnosed during their stay in ICU. This involved all patients in whom oral antihyperglycemic agents and/or (non-)insulin injectable drugs were prescribed immediately following ICU stay, as well as patients who underwent major pancreatic surgery.

We then used a nested case-control design among 1-year ICU survivors who responded to a follow-up questionnaire. To this end, information about survival status and current place of residence was retrieved from the Dutch municipality registers. If a patient was still alive, a follow-up questionnaire was sent per postal mail approximately 14 months after ICU discharge. The Medical Ethics Committee of the University Medical Center Utrecht determined that the current study did not fall under the Medical Research Involving Human Subjects Act (IRB number 10-005).

Data collection

Approximately one year after ICU discharge, study patients (or their relatives) responded to a routine follow-up questionnaire that included, among others, queries about current medication use, body weight, and physical functioning (i.e., Barthel Index of Activities of Daily Living) (22). Demographic data, medical history, and information about the ICU stay were retrieved from electronic health records. Hba1c concentration was available for a subgroup of consecutive patients, as it was systematically measured upon ICU admission without specific indication during a restricted period of time (February 2011 – October 2013).

Definition of cases and controls

Subjects were classified as cases if they used oral antihyperglycaemic agents (including metformin, glimepiride, gliclazide, and others), insulin or non-insulin injectable diabetes medications, or both at the time of follow-up, and had not used such medications prior to ICU admission. All other responders to the questionnaire without DM were eligible as potential controls. For each case, two control subjects were randomly selected after matching on age (plus or minus five years), sex, overweight (body mass index (BMI) >25 kg/m^2), and solid-organ transplant as reason for ICU admission, since these factors might act as either confounders or effect modifiers of the association under study (23-25). Subsequently, in-depth case-reviews using electronic health records were performed in all subjects to verify absence of clinically recognized DM2 and/or prior pharmacological treatment with glucose-lowering drugs.

Definition of exposure variables

Hyperinflammation and tissue hypoxia were the exposures of primary interest (Figure 1). Hyperinflammation was parameterized as the number of days with C-reactive protein (CRP) levels >100 mg/L occurring before day 4 in the ICU. This cut-off was chosen based on the median of observed CRP values. Likewise, the burden of tissue hypoxia was parameterized as the number of days with serum lactate levels >2 mmol/L occurring before day 4 in the ICU. This cut-off was derived from the sepsis-3 definition for shock (26). When missing, CRP and lactate levels were assumed to be in the lower range (respectively <100 mg/L and <2 mmol/L). We decided to study these specific markers because CRP was daily measured in almost all patients and lactatemia was unlikely to be missed because of low-threshold use of lactate measurement in case of any suspicion. Other potential risk factors for diabetes onset that were examined include use of corticosteroids during ICU admission, caloric intake and (par)enteral feeding, and post-discharge factors such as weight change, medication use, and physical functioning. Additionally, critical illness-induced hyperglycaemia was considered a proxy of possible etiological factors on the causal pathway leading to DM2. It was categorized as either mild or severe based on blood glucose levels between 7.9 and 11.1 or ≥11.1 mmol/L measured at any time in ICU, respectively.

Statistical analysis

Potential risk factors for new-onset DM2 were first compared between cases and controls using explorative univariable analyses. Based on the causal model shown in Figure 1, we then more formally assessed the association between hyperinflammation and/or tissue hypoxia during critical illness and the occurrence of new-onset DM2 in the first year after discharge using multivariable logistic regression analysis. We also performed a stratified analysis for patients admitted after solid organ transplantation, as transplant status is a strong risk factor for the development of DM2 and might be an effect modifier (27, 28). Finally, a sensitivity analysis was performed in patients having an ICU length of stay of more than 2 days. We used unconditional logistic regression as we believe that our matching procedure was unlikely to introduce sparse data. Furthermore, using unconditional logistic regression enables the possibility to perform a stratified analysis (29). To address any residual confounding not fully accounted for by the matching procedure, the analysis also included age and BMI as covariables. Although cases and controls were matched on these factors, some variability remained as matching occurred within age ranges and using a single BMI cut-off (30).

Categorical and continuous data were compared between groups using Wilcoxon Rank-Sum tests or Chi-square tests, as appropriate. All analyses were performed in SAS Enterprise Guide 4.3 (SAS Institute, Cary, NC) and R Studio (R Studio Team 2015, Boston, MA).

Results

Incidence of new-onset DM2

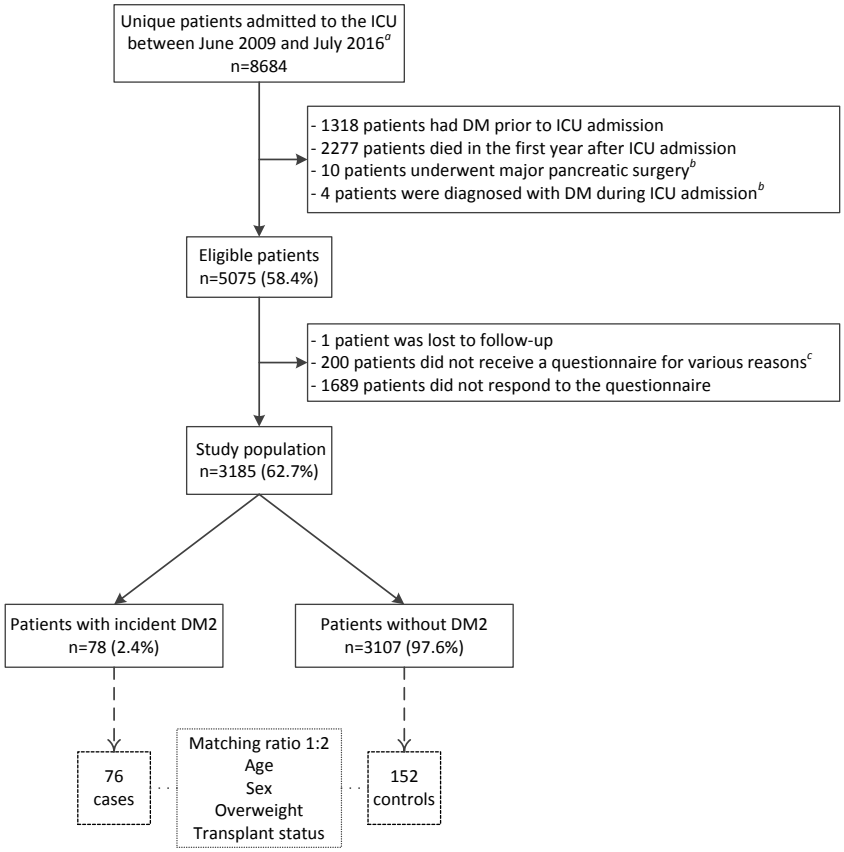
Between June 2009 and July 2016, a total of 8684 patients were treated in our ICU (not including patients after uncomplicated elective cardiothoracic surgery). Patient inclusion is shown in Figure 2.

Among the 5075 patients who were eligible for study inclusion, 4874 were sent a questionnaire. Response data were available for 63%, yielding a final study population of 3185 patients. New-onset DM2 occurred in 78 of these 3185 patients, corresponding to a cumulative incidence of new-onset DM2 of 2.4% (95%CI 2.0-3.0%) over a follow-up period of median 426 (IQR 405 to 446) days. Cases were identified based on newly-initiated treatment with oral antihyperglycaemic agents (n=62; 79%), (non-)insulin subcutaneous drugs (n=12; 16%), or both (n=4; 5%) since ICU discharge. Of these 78 patients, 76 cases could be matched to 152 controls.

Descriptive analyses

Patient demographics, comorbidities, characteristics of the ICU-stay, and post-discharge patient factors (measured at follow-up) are listed in Table 1.

Figure 2. Flowchart of patient inclusion



DM: diabetes mellitus, ICU: intensive care unit. * Not including patients after elective and uncomplicated cardiothoracic surgery. ** These criteria were applied during the final selection of cases and controls. *** Including, but not limited to, not willing to participate or living abroad.

Cases were more severely ill upon admission to the ICU and were more frequently admitted for medical reasons and/or sepsis than controls. In contrast, no statistically significant differences were observed between cases and controls regarding neither total caloric intake, nor the use of parenteral nutrition or corticosteroids during ICU admission. Median ICU length of stay was slightly longer in cases than controls, but this difference did not reach statistical significance.

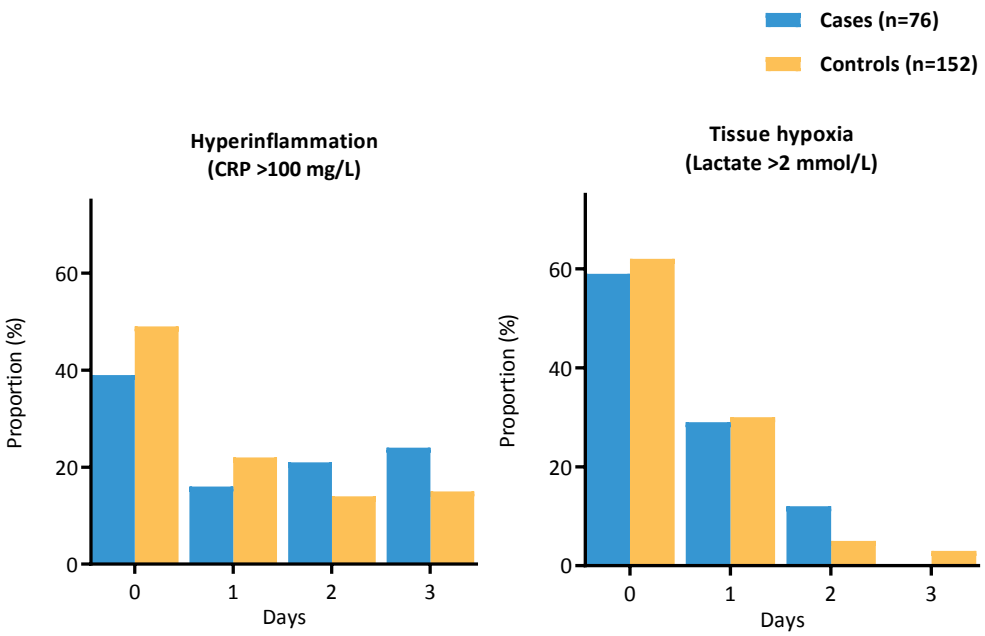
During the first 24 hours of ICU admission, cases had higher median (IQR) blood glucose levels than controls (respectively 10.9 (9.3-12.9) versus 9.8 (8.2-11.6) mmol/L; p=0.010). Furthermore, episodes of severe hyperglycaemia were more frequently observed in cases than controls (p=0.016), despite the fact that 76% and 64% of patients received insulin in both groups (p=0.070). Of note, median insulin doses were higher in cases (47 (IQR 26-75) versus 35 (17-57) IU per day; p=0.011). Although all study patients

Table 1. Patient, illness, and post-discharge characteristics of cases and controls (n=228)

Factors	Cases (n=76)	Controls (n=152)	P-value
Patient factors			
Age (years)	59 (51-67)	60 (52-67)	MF
Female gender	34 (45)	68 (45)	MF
Body mass index (Kg/m^2)	27 (24-30)	27 (25-29)	0.504
Overweight[a]	55 (72)	110 (72)	MF
Charlson comorbidity index	1 (0-2)	1 (0-2)	0.490
Hypertension	23 (30)	49 (32)	0.763
Current smoking	3 (4)	9 (6)	0.755
Use of lipid lowering drugs	19 (25)	35 (23)	0.741
Acute disease factors			
APACHE IV score	60 (47-79)[g]	54 (41-69)[h]	0.037
Admission for solid organ transplant	21 (28)	42 (28)	MF
Recent surgery prior to ICU admission[b]	50 (66)	122 (80)	0.017
Sepsis[c]	23 (30)	26 (17)	0.023
Maximum CRP (mg/L)	125 (74-218)[h]	101 (56-160)[h]	0.043
Days with CRP >100 mg/L[d]	1 (0-2)	1 (0-2)	0.048
Maximum lactate (mmol/L)	2.7 (2.0-4.2)[i]	2.8 (1.5-4.3)[i]	0.679
Days with lactate >2 mmol/L[d]	0 (0-1)	0 (0-1)	0.523
Blood glucose level upon ICU admission	10.9 (9.3-12.9)[h]	9.8 (8.2-11.6)	0.010
Hyperglycaemia[e]			0.016
- Mild	23 (30)[h]	62 (41)	
- Severe	47 (62)	68 (45)	
Use of insulin	58 (76)	97 (64)	0.070
Use of vasopressors	51 (67)	98 (64)	0.694
Use of corticosteroids	36 (47)	61 (40)	0.298
(Par)enteral nutrition	47 (62)	89 (59)	0.633
Mean daily caloric intake (kcal)	1206 (345-1642)[h]	966 (149-1524)[h]	0.151
ICU length of stay (days)	3 (1-7)	2 (1-5)	0.058
Post-discharge factors			
Weight change since ICU admission (kg)	−1 (−6-6)[j]	−3 (−7-3)[j]	0.136
Chronic use of tacrolimus	24 (32)	41 (31)	0.882
Chronic use of oral corticosteroids	30 (39)	37 (24)	0.018
Dependency in physical functioning[f]	12 (33)[k]	19 (31)[k]	0.804

Continuous variables are presented as median (IQR), dichotomous/categorical variables are presented as frequency (%). MF: matching factor, APACHE: acute physiology and chronic health evaluation, ICU: intensive care unit, CRP: C-reactive protein. [a] Overweight was defined as a body mass index >25 Kg/m^2. [b] Recent surgery prior to ICU admission was defined as elective or emergency surgery in the 24 hours prior to ICU admission. [c] Sepsis was defined according to sepsis-3 definitions. [d] When CRP or lactate was not measured, it was assumed to be normal (i.e., <100 mg/L for CRP and <2 mmol/L for lactate). [e] Scored 'mild' or 'severe' when at least a single blood glucose level measurement was between 7.9 and 11.1 or ≥11.1 mmol/L, respectively. [f] A score of 10 or higher on the Barthel Index of Activities of Daily Living, only included in the questionnaire for patients admitted in 2012 or later with a minimal length of stay of 48 hours (47% of cases and 41% of controls). [g] ≤10% missing values, [h] ≤5% missing values, [i] ≤50% missing values, [j] ≤20% missing values, [k] ≤60 missing values.

Figure 3. Occurrence of hyperinflammation and tissue hypoxia during the first 4 days in ICU (n=228)



did not have clinically recognized DM prior to ICU admission, we observed evidence of pre-existing disturbances of glucose metabolism in some patients as indicated by elevated HbA1c values. Among the 26 cases and 53 controls for whom this biomarker was available, median (IQR) HbA1c concentrations were 43.0 (40.0-50.0) and 40.0 (37.0-43.0) mmol/mol, respectively (p=0.004). In fact, there were 7 cases and 3 controls in whom baseline concentrations were higher than the 48 mmol/mol diagnostic cutoff-level recommended by the American Diabetes Association (31).

One year after ICU discharge**,** there were no statistically significant differences in weight change, use of tacrolimus, or physical impairments. However, use of corticosteroids was more frequent in cases than controls (39% versus 24%, respectively; p=0.018).

Burden of hyperinflammation and tissue hypoxia

Figure 3 shows the occurrence of hyperinflammation and tissue hypoxia during the first 4 days of ICU admission.

Median CRP values were 125 (IQR 74-218) and 101 (56-160) mg/L in cases and controls, respectively (p=0.043). Cases also had more days with CRP >100 mg/L during their first 4 days in ICU (median 1.0 (IQR 0-2.0) versus 1.0 (0-2.0) days; p=0.048). In contrast, we observed similar days with lactate >2 mmol/L (median 0 (IQR 0-1)) in both cases and controls (p=0.523).

Table 2. Multivariable logistic regression analyses of the association between hyperinflammation, tissue hypoxia and new-onset diabetes mellitus

Type of analysis	Patients	Cases (n)	Controls (n)	OR	95%CI	P-value
Primary analysis	**All cases and controls**	76	152			
	Hyperinflammation			1.98	1.10 – 3.55	0.022
	Tissue hypoxia			1.02	0.57 – 1.84	0.936
Sensitivity analysis	**ICU length of stay of ≥2 days**	44	76			
	Hyperinflammation			2.68	1.18 – 6.09	0.019
	Tissue hypoxia			1.18	0.55 – 2.55	0.671
Stratified analysis	**Solid organ transplantation**	21	42			
	Hyperinflammation			1.00	0.34 – 2.96	0.860
	Tissue hypoxia			1.10	0.38 – 3.23	0.997
	Other diagnoses[a]	55	110			
	Hyperinflammation			2.76	1.32 – 5.75	0.007
	Tissue hypoxia			1.03	0.49 – 2.17	0.928

Although cases and controls were matched on age (range ±5 years) and overweight (BMI >25 Kg/m^2), the analysis was adjusted for age in years and body mass index to remove residual confounding by these variables. OR: odds ratio, ICU: intensive care unit. Hyperinflammation was defined as ≥2 days CRP >100 mg/L in the first 4 days of ICU admission. Tissue hypoxia was defined as ≥1 day lactate >2 mmol/L in the first 4 days of ICU admission. [a] Including malignancies (~20%), cardiology or cardiothoracic surgery (~20%), neurology or neurosurgery (~15%), sepsis (~10%), trauma (~10%), and other admission diagnosis (~25%).

In multivariable logistic regression analysis, the odds for developing new-onset DM2 were higher in patients exposed to hyperinflammation (OR 1.98, 95%CI 1.10-3.55; p=0.022), but not tissue hypoxia (OR 1.02 (95%CI 0.57-1.84; p=0.936) (Table 2). These associations remained in a sensitivity analysis including only the 44 cases and 76 controls having an ICU length of stay ≥2 days. Of note, in this specific sensitivity analysis the performed matching was broken and, therefore, the results are more sensitive to confounding. In a third analysis following stratification upon transplant status, the association between hyperinflammation and new-onset DM2 was not observed among solid organ transplant recipients, yet became stronger in remaining patients, suggesting that the pathophysiological mechanisms leading to diabetes may be different in these two groups.

Discussion

The main finding of our study is that prolonged exposure to hyperinflammation during critical illness was independently associated with the subsequent development of DM2. This confirms our hypothesis that severe inflammation might not only aggravate short-

term critical illness-induced hyperglycaemia but may also be causally related to new-onset DM2. In contrast, oxidative stress-induced beta cell injury seems only a minor factor in the development of DM2 in critically ill patients. Furthermore, we observed a 2.4% (95%CI 2.0-3.0%) cumulative incidence of DM2 in the first year after ICU discharge, which is substantially higher than the approximately 1% annual rate reported for an age and gender matched Dutch population (32), and confirms previous estimates of increased DM2 occurrence following critical illness (14, 15).

A pathophysiological pathway might be ongoing low-grade inflammation following critical illness. This phenomenon has been reported to affect long-term outcomes in ICU survivors (33, 34). In addition, several prospective studies have reported that inflammation is independently associated with incident DM2 in middle-aged adults in several settings (9, 35-37). It might reflect the influence of inflammation on endothelial function altering permeability and peripheral blood flow which might limit insulin delivery (9, 36).

Another potential mechanism underlying the observed association might be that several biomarkers playing a role in the inflammatory processes during critical illness (e.g., IL-1, TNF-α, and CRP) induce dysfunction or dead of B-cells leading to permanent damage of pancreatic islets (35, 38, 39). As higher age is associated with insulin resistance, it might well be that a part of our study population already had a decline of insulin action when compared to younger patients (40). Normally, this does not directly cause problems due to the ability of the body to compensate for insulin resistance through B-cell hyperplasia and increased insulin secretion (38, 39). A loss of B-cells due to acute disease, though, might disturb this balance leading to overt DM2 after ICU discharge. This mechanism fits the observation of high HbA1c levels in some patients in our cohort suggesting that some of the cases and controls in our study had unrecognized preclinical DM2 at ICU admission, but that islet dysfunction progressed afterwards necessitating medical treatment with oral antihyperglycaemic agents and/or (non-)insulin injectables.

Interestingly, hyperinflammation was not associated with new-onset DM2 in our stratified analysis of post-transplant patients, although the overall incidence was highest in this subgroup. Indeed, an immunosuppressed state appears to be the most important risk factor for post-transplant DM2, possibly negating any inflammatory effects. A systematic review of 19 studies reported that the type of immunosuppressive regimen (including steroids, cyclosporine, and tacrolimus) explained 74% of the incidence of DM2 in this population (27). Therefore, the chance to detect any other mechanism in a small subgroup population, such as in this study, is unlikely.

For this study, we used a large cohort of patients admitted to our ICU over a period of more than 5 years, allowing estimation of an absolute incidence proportion. We used a very pragmatic definition of new-onset DM2, which was solely based on initiation of new pharmacological treatment to achieve glycemic control. Furthermore, case detection

relied on self-reported medication use. Both methods may result in misclassification, most likely leading to underestimation of true DM2 incidence and probably a dilution of the observed effect. On the other hand, as an ICU admission provokes subsequent contact with several health care professionals, the chance to be diagnosed with DM2 during follow-up might be increased. However, the incidence proportion that we found is comparable to studies in similar populations, supporting the reliability of our results (14, 15). In addition, non-response to the follow-up questionnaire might be associated with risk factors for new-onset DM2, thereby introducing selection bias. We used only CRP and lactate levels as a proxy of hyperinflammation and tissue hypoxia, respectively. It is arguable whether this is indeed sufficient to investigate a possible etiologic association. We recommend that future research should include multiple constructs to operationalize hyperinflammation and tissue hypoxia. Last, we aimed to answer an etiologic question using a case-control design, but our study was hampered by the competing risks of both loss-to-follow-up and death. In addition, due to a limited number of cases we were not able to adjust for all potential confounding variables. Therefore, the results of our study have to be interpreted with caution and large prospective studies are needed to confirm our hypothesis.

Conclusions

Patients with severe inflammation during critical illness have an increased risk to develop DM2 after ICU discharge. Tissue hypoxia was not associated with new-onset DM2. Follow-up of this particular risk group might enable physicians to prevent the progression of peripheral insulin resistance and pancreatic islet dysfunction, for instance by promoting lifestyle changes, and to start appropriate treatment in an early phase to prevent disease-associated complications.

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6

Chapter 6

Occurrence and risk factors of chronic pain after critical illness

Maria E. Koster-Brouwer
Mienke Rijsdijk
Wouter K.M. van Os
Ivo W. Soliman
Arjen J.C. Slooter
Dylan W. de Lange
Diederik van Dijk
Marc J.M. Bonten
Olaf L. Cremer

In submission

Abstract

Purpose Occurrence, risk factors, and impact on daily life of chronic pain after critical illness have not been systematically studied.

Methods We surveyed patients who had been discharged from our tertiary Intensive Care Unit (ICU) between 2013 and 2016. Three cohorts were defined: (1) ICU survivors; (2) one-year survivors reporting newly-acquired chronic pain; (3) one-year survivors with pain who lived within 50 kilometer from the study hospital. In cohort 1 we estimated the prevalence of new chronic pain one year after ICU discharge and constructed a prediction model for its occurrence incorporating three outcomes: death during follow-up, surviving without new pain, and surviving with newly-acquired pain. In cohort 2 we determined clinical features of pain and its impact on daily life. In cohort 3 we assessed the presence of neuropathic characteristics of pain.

Results The three cohorts contained 1842, 160, and 42 patients, respectively. Estimated occurrence of new chronic pain was 17.7% (95%CI 15.8-19.8%; n=242) in one-year survivors (n=1368). Median pain intensity on the numeric rating scale was 4 (IQR 2-6) in the week before survey response, with impact being most evident on activities of daily living, social activities, and mobility. Neuropathic pain features were present in 50% (95%CI 37-68%) of affected subjects. Among nine predictor variables included in a multinomial model, only female gender and days in ICU with hyperinflammation were associated with pain.

Conclusions Newly-acquired chronic pain is a frequent consequence of critical illness, and its impact on daily life of affected patients is substantial.

Introduction

Over the past decades, the main focus of outcomes research after intensive care unit (ICU) admission has shifted from short-term mortality towards longer-term patient-centered outcomes, such as physical disability, cognitive dysfunction, and impaired mental health. It is now well acknowledged that a substantial number of ICU survivors experience disabilities in one or more of these areas, which collectively have been coined as the post-intensive care syndrome (1). An important late consequence of critical illness that may contribute to physical disability and reduced health-related quality of life (HRQoL) is chronic pain (2, 3). Pain already affects many individuals in the general population, and it is expected to become an even larger health problem in the near future (4, 5). In addition to having detrimental impact on HRQoL in affected individuals, chronic pain also has major consequences for society at large, causing high healthcare costs and work absenteeism (6).

At present, only few studies have reported the occurrence of chronic pain after critical illness. These were generally hampered by small sample sizes, high rates of loss to follow-up, and use of questionnaires that were not primarily intended to assess pain (i.e., pain information was mostly derived from subdomains within HRQoL questionnaires) (3, 7). As a consequence, there is also a lack of information about the clinical characteristics of chronic pain after critical illness. This information may help categorize pain into nociceptive, neuropathic, or mixed pain, and provide important insight into possible underlying pathophysiological mechanisms, which could then guide development of preventive or preemptive treatment (8). Furthermore, identification of risk factors for the development of chronic pain after ICU discharge is required to target such strategies at individuals who are at highest risk.

The aim of our study was to estimate the prevalence of newly-acquired chronic pain one year after critical illness and to describe its severity, characteristics, and impact on HRQoL and activities of daily living (ADL). Additionally, we developed a prediction model for its occurrence.

Methods

Study population and design

We conducted a cohort study among adult patients admitted to the mixed ICU of the University Medical Center Utrecht in the Netherlands between January 2013 and December 2016. To be included in the study, patients had to be admitted to the ICU for a minimal duration of 48 hours and needed to be discharged alive. For patients with multiple ICU admissions, only the first ICU admission was included.

Based on the overall cohort, we constructed three datasets; the first containing all ICU survivors fulfilling the eligibility criteria *(dataset 1)*, the second including patients who had survived for at least one year after ICU discharge, responded to a follow-up questionnaire, and reported to have newly-acquired chronic pain *(dataset 2)*, and the third including one-year survivors who reported pain and lived within a 50 kilometer radius from the study hospital *(dataset 3)*. We used these datasets to estimate the prevalence of newly-acquired chronic pain one year after ICU discharge in patients being alive at that moment (dataset 1), describe its clinical features and impact on HRQoL and daily life (dataset 2), and prospectively assess the presence of nociceptive or neuropathic characteristics (dataset 3). Similarly, the derivation of a prediction model for the occurrence of new chronic pain after ICU discharge was based on dataset 1.

The medical ethics committee of the UMCU concluded that the retrospective part of the study (datasets 1 and 2) did not fall within the ambit of the Medical Research Involving Human Subjects Act (IRB-number 10-006), whereas for the prospective substudy among patients reporting chronic pain (dataset 3) written informed consent was obtained (IRB-number 17-022).

Data collection and study outcomes

All clinical data were retrieved from electronic health records. Survival status was derived from the Dutch municipal personal records database. Patients who were alive one year after ICU discharge received a survey by postal mail, including specific queries on newly-acquired pain (e.g. presence, location, intensity, interference with daily life) and current drug use.

The primary outcome of our study was self-reported newly-acquired chronic pain. This was defined as pain that had developed or worsened since ICU admission and remained still present one year after discharge. Reported pain characteristics included intensity (expressed as a numeric rating scale (NRS)), body location, self-perceived impact on daily living, and use of medications for pain relief. To construct a robust outcome measure (i.e., resilient to reporting errors), patients needed to have a positive response on at least two (unrelated) survey items indicating presence of newly-acquired pain.

To identify risk factors for the development of chronic pain, we *a priori* selected predictor variables based on literature review (7, 10-13) and clinical expertise. These included age, gender, admission type, ICU length of stay; presence of several comorbid conditions, obesity, surgical wounds, and decubitus ulcers; duration of mechanical ventilation, organ failure, hyperinflammation, pain, delirium, sedation, and analgesic drug use. For these latter variables we considered the first 14 days in ICU only in order to reduce potential co-linearity with total ICU length of stay. Parameterization of predictor variables is included in Appendix I).

Additional follow-up of patients included in dataset 3 was obtained by performing home visits. To asses the presence or absence of neuropathic pain characteristics we used the validated Dutch version of the Douleur Neuropathique 4 (DN4) questionnaire at a previously defined cut-off value >4 (14-16). Detailed information about this examination is provided in the supplementary material (Appendix II).

Statistical analyses

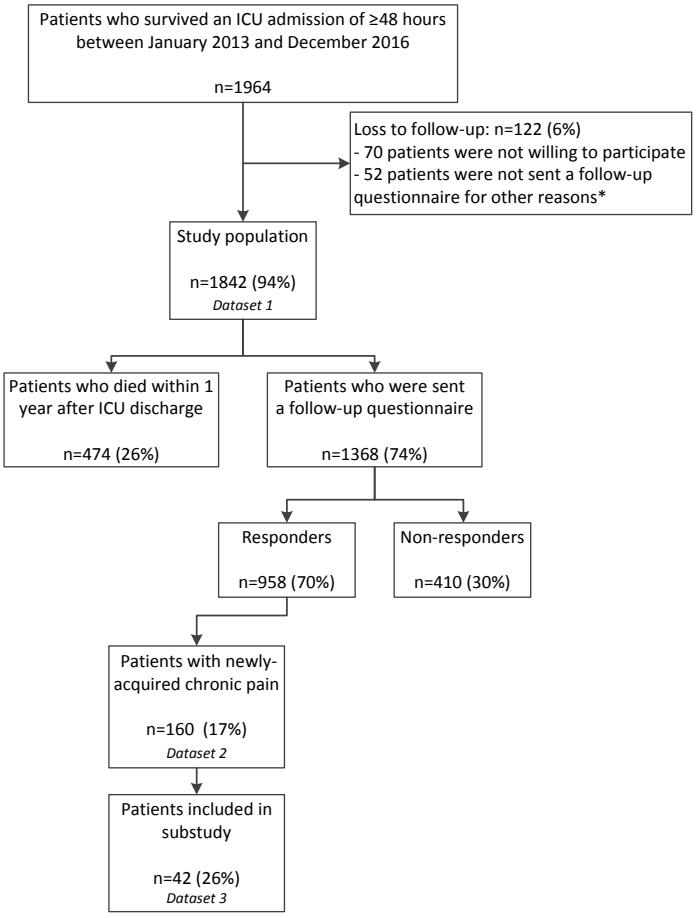
Using dataset 1, we developed a prediction model for the occurrence of newly-acquired pain after one year, using information available at the moment of ICU discharge. To minimize bias by selective loss to follow-up, we used multiple imputation techniques to impute the outcome for patients who did not respond to the questionnaire. In this procedure, 30 imputed datasets were created as the amount of non-response was approximately 30%. In addition, we developed decision rules to handle missing data in predictor variables. A description of the imputation procedure and decision rules is provided in the supplementary material (Appendix III).

Subsequently, we derived a multinomial logistic regression model to estimate probabilities of survival without new chronic pain, survival with newly-acquired chronic pain, and death. The latter was incorporated as an outcome because it is a competing event. For all candidate predictors measured on a continuous scale, it was evaluated whether using cubic splines would result in a better model fit than did modelling a linear association (17). Predictor selection was based on the least absolute selection and shrinkage operator (LASSO) (18, 19). After applying the LASSO-procedure to each imputed dataset, we used a majority vote for final model specification (i.e., a predictor was selected if it remained in the model in more than half of the imputed datasets).

Model performance was evaluated using the area under the curve (AUC) for dichotomized comparisons across the three outcomes, the most important being newly-acquired chronic pain versus a composite of both other categories. Hosmer-Lemeshow goodness-of-fit tests (with accompanying calibration plots) were conducted similarly. Finally, a correct classification rate was estimated using the observed prevalence of newly-acquired chronic pain as a cut-off for categorization of the predicted probabilities.

Continuous data are presented as medians with interquartile ranges, categorical data as absolute numbers with percentages. Differences between subgroups of patients were assessed using Mann-Whitney U, Chi-square, or Kruskal-Wallis tests, as appropriate. For all analyses, p-values <0.05 were considered statistically significant. When applicable, multiple analyses were pooled using Rubin's rules (20, 21). All analyses were performed using SAS Enterprise Guide 4.3 (SAS Institute, Cary, NC) or Rstudio (RStudio Team (2015). RStudio: Integrated Development for R. RStudio, Inc., Boston, MA).

Figure 1. Flowchart of patient inclusion



ICU: intensive care unit. * Including, but not limited to, no information on address or survival status available in the municipal registries.

Results

Study cohort

Among 1964 ICU survivors who had been admitted for at least 48 hours between January 2013 and December 2016, 122 patients were lost to follow-up. The remaining 1842 subjects were included in our study (Figure 1). Of these, 474 (26%) died in the first year after ICU discharge, and 1368 received a follow-up questionnaire, to which 958 (70%) responded. The median (IQR) time to follow-up in these subjects was 439 (419-461) days (i.e., approximately 14 months).

Occurrence and characteristics of newly-acquired chronic pain

The proportion of one-year survivors (dataset 1) developing newly-acquired chronic pain was estimated to be 17.7% (95%CI 15.8-19.8%; n=242). Patients acquiring chronic pain during follow-up were slightly younger, more often female, and had more frequently used opioids prior to ICU admission than other patients (Table 1). Affected patients were also exposed to more days with hyperinflammation, mechanical ventilation, and high-dose opioid use during the first two weeks in ICU. Descriptive characteristics of patients who died during follow-up are provided in the supplementary material (Appendix IV, Table S.1.).

Among the 160 responders to the survey who reported newly-acquired chronic pain (dataset 2), the median pain intensity during the last week before survey response was 4 (IQR 2-6) on the NRS (Figure 2). Of note, 10% of subjects reported an average pain intensity of 0, suggesting that they experienced pain attacks instead of having continuous pain. Body regions most affected by pain were the thorax (32%), legs and knees (29%), feet (26%), shoulders (25%), and the back (21%) (Figure 3). Pain affecting more than one body region was reported by 68% of patients. Use of opioids and/or antineuropathic drugs at follow-up was reported by 22 (14%) and 10 (6%) patients, respectively, whereas 3 patients used both. Behavioral impact of pain was most evident in the domains of ADL, social activities, and mobility (Figure 2).

Nociceptive versus neuropathic features

Among 87 patients eligible for additional follow-up, 6 subjects were unable or unwilling to participate. The median time to follow-up in this subgroup was 35 (27-44) months since ICU discharge. Of note, at this point in time 39 (45%) subjects reported to have recovered from the pain they had previously reported. Characteristics of the 42 remaining patients (dataset 3) are presented in the supplementary material (Appendix V, Table S.2.). Based on a DN4 score ≥4, we considered pain to be predominantly of neuropathic origin in 21 (50%; 95%CI 37-68%) of these patients. Among them, 12 (57%) reported multiple body sites to be affected, compared to 9 (43%) of their counterparts who experienced mainly nociceptive pain (p=0.876). Furthermore, pain intensity during the last week prior to the home visit was significantly higher in patients having neuropathic compared to nociceptive pain (median VAS 5 (IQR 3-6) versus 3 (2-5); p=0.036, and NRS 6 (4-7) versus 4 (3-6); p=0.030, respectively).

Predicting newly-acquired chronic pain

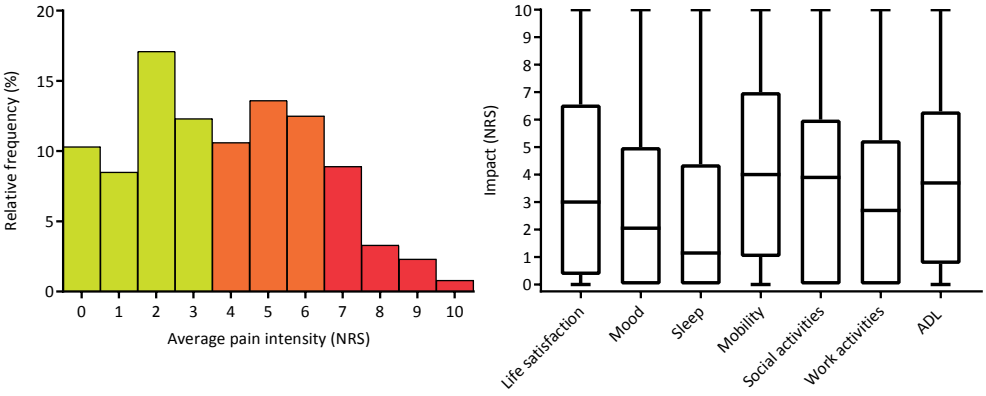
Univariable comparisons for all *a priori* selected predictor variables are reported for survivors with and without pain in Table 1, and for survivors and non-survivors in Table S.1 (Appendix IV). Background probabilities of survival without new chronic pain, survival with newly-acquired chronic pain, and death were 61.1%, 13.1%, and 25.7%,

Table 1. Demographics and characteristics of ICU stay in patients with and without newly-acquired chronic pain (n=1368)

	Patients without newly-acquired chronic pain	Patients with newly-acquired chronic pain	
	n=1126	n=242	P-value
Age, years	59 (47-69)	56 (43-66)	0.034
Gender, male	751 (66.7)	129 (53.3)	<0.001
Comorbidities			
- Diabetes mellitus	157 (13.9)	33 (13.6)	0.713
- Any malignancy	182 (16.1)	30 (12.6)	0.259
- Neurocognitive deficit[a]	170 (15.1)	37 (15.1)	0.803
- Connective tissue disease	40 (3.6)	8 (3.3)	0.845
- Obesity	244 (21.7)	51 (21.1)	0.707
Drug use before ICU admission			
- Opioids	65 (5.7)	25 (10.5)	0.035
- Antineuropathic drugs	26 (2.3)	8 (3.4)	0.521
APACHE IV score	70 (55-87)	69 (53-87)	0.606
Admission type			0.227
- Medical	550 (48.8)	117 (48.4)	
- Surgical elective	280 (24.9)	50 (20.5)	
- Surgical non-elective	296 (26.3)	75 (31)	
Surgical procedure during ICU stay	351 (31.2)	90 (37.1)	0.126
Pressure ulcers[b]	193 (17.2)	44 (18.1)	0.692
Number of days with			
- Hyperinflammation (CRP > 100 mg/L)	2 (1-5)	3 (2-6)	0.002
- Organ failure (SOFA > 5)	2 (0-4)	2 (0-4)	0.612
- Mechanical ventilation	4 (2-8)	5 (2-10)	0.028
- Sedation (RASS < -2)	1 (0-4)	1 (0-4)	0.129
- Deliriumc	0 (0-2)	0 (0-2)	0.385
- Pain (NRS > 3)d	1 (0-2)	1 (0-2)	0.127
- Opioid use[e]			
- Low dose	2 (1-4)	2 (1-5)	0.530
- Intermediate dose	2 (1-3)	2 (1-3)	0.636
- High dose	1 (0-2)	1 (0-3)	0.049
Other analgesic drug use			
- Paracetamol/NSAID	1089 (96.7)	237 (98.1)	0.363
- Antineuropathic drugs	104 (9.2)	31 (12.8)	0.157
- Alpha-2 agonists	533 (47.4)	121 (49.9)	0.559
- Epidural or locoregional analgesia	71 (6.3)	19 (8)	0.490
ICU length of stay, days	6 (3-11)	6 (4-12)	0.119

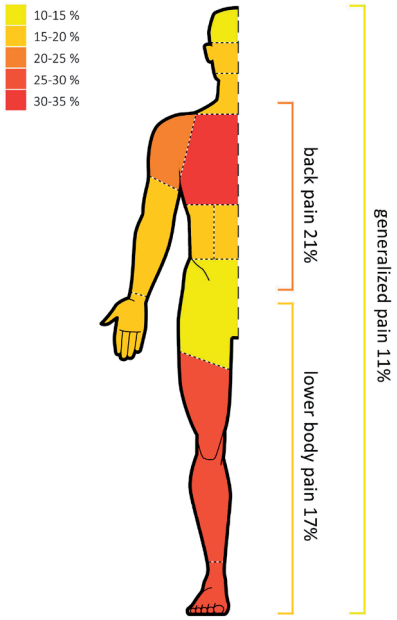
APACHE: acute physiology and chronic health evaluation, ICU: intensive care unit, CRP: C-reactive protein, SOFA: sequential organ failure assessment, RASS: Richmond agitation-sedation scale, NRS: numeric rating scale, NSAID: nonsteroidal anti-inflammatory drugs. Continuous and categorical data are presented as medians (first – third quartile) and frequency (percentage), respectively. [a] Cerebrovascular disease, depression, and/or dementia. [b] Stage III or IV, according to the National Pressure Ulcer Advisory Panel (NPUAP) guidelines. [c] Daily assessed using the confusion assessment method for the intensive care unit (CAM-ICU) in patients who were not sedated. [d] Daily assessed using a numeric rating scale in patients who were not sedated. [e] Days with low, intermediate and high dose opioids were defined as <30, 30-60, and ≥60 mg oral morphine equivalents per 24 hours, respectively.

Figure 2. Pain intensity during last week prior to assessment and its self-perceived impact on activities of daily life in responders reporting newly-acquired chronic pain (n=160) at follow-up



NRS: numeric rating scale. ADL: activities of daily living.

Figure 3. Affected body locations in responders reporting newly-acquired chronic pain (n=160) at follow-up



respectively. Using LASSO, nine predictor variables were selected for inclusion in the multinomial logistic regression model (Table 2).

Female gender and days with hyperinflammation were most predictive for the development of chronic pain after critical illness, whereas the other variables were

Table 2. Predictor variables for newly-acquired chronic pain after critical illness

	Coefficient	OR (95% CI)	P-value
Intercept	-1.271	-	0.061
Age, years[a]	0.002	1.00 (0.97-1.03)	0.912
- Age, years′	-0.018	0.98 (0.93-1.04)	0.570
- Age, years′′	0.087	1.09 (0.68-1.75)	0.749
Gender, male	-0.634	0.53 (0.40-0.71)	<0.001
Any malignancy	-0.300	0.74 (0.48-1.13)	0.222
APACHE IV score	-0.002	1.00 (0.99-1.00)	0.659
Days with hyperinflammation[b]	0.086	1.09 (1.03-1.15)	0.009
Days with sedation[c]	0.005	1.00 (0.95-1.06)	0.886
Days with pain[d]	0.047	1.05 (0.96-1.14)	0.357
Days with high dose opioids[e]	-0.034	0.97 (0.90-1.04)	0.390
ICU length of stay, days	0.004	1.00 (0.99-1.02)	0.512

Regression coefficients were estimated using a multivariable multinomial logistic regression model that included death, surviving with newly-acquired chronic pain, and surviving without pain one year after ICU discharge as discrete outcome categories. Predictor selection was based on the least absolute selection and shrinkage operator (LASSO). Only coefficients for newly acquired chronic pain after critical illness are shown here; coefficients for death during follow-up are reported in Table S.3. APACHE: acute physiology and chronic health evaluation, ICU: intensive care unit. For all 'days with' the first 14 days of ICU admission are taken into account. [a] Modelled as a restricted cubic spline with 4 knots, namely 25, 55, 67, and 80 years. [b] C-reactive protein > 100 mg/L, [c] Richmond agitation sedation-scale < -2, [d] Numeric rating scale > 3, [e] ≥ 60 mg oral morphine equivalents per 24 hours.

mainly associated with death (Appendix VI, Table S.3.). Discriminative ability for death was acceptable (AUC 0.739; 95%CI 0.644-0.816), yet limited for predicting the occurrence of new chronic pain (AUC 0.666; 95%CI 0.565-0.754). Calibration plots for both (dichotomized) outcomes are shown in the supplementary material (Appendix VII, Figure S.1.), and Hosmer-Lemeshow tests indicated these were acceptable (p=0.903 and p=0.311, respectively).

Overall, the model yielded predicted probabilities for acquiring chronic pain after ICU discharge that ranged between 0.019 and 0.434 in individual patients (median 0.133 (IQR 0.088-0.171)). For subjects with newly-acquired chronic pain, without newly-acquired chronic pain, and those who had died, these probabilities were 0.161 (0.120-0.202), 0.139 (0.097-0.178), and 0.093 (0.070-0.139), respectively. Using a >13% predicted risk (i.e., the prior probability based on the background probability for newly-acquired chronic pain) as a cut-off for classification, the model yielded a positive predictive value of 18% and a negative predictive value of 92%. This implies that 69% of survivors with newly-acquired pain, 43% of survivors without (new) pain, and 73% of deceased patients in our dataset could be correctly predicted by the model.

Discussion

Based on a study population of 1368 one-year survivors, we estimated that 18% (95%CI 16-20%) developed chronic pain during the first year after ICU discharge. Pain intensity was considerable, involved a wide range of body regions, and significantly impacted the life of affected patients. Furthermore, features that suggested underlying neuropathy were observed in approximately half of patients who could be prospectively evaluated in greater detail. Although there were a number of clinical factors related to the development of chronic pain, possibilities to accurately predict its occurrence seem limited.

Based on studies in the general population, the prevalence of chronic pain in the Netherlands is known to be 18% (6, 22). Compared to this background, a 18% rate of new cases occurring during the first year after ICU discharge is substantial. As the number of ICU survivors is expected to rise in the future (23, 24), the prevalence of chronic pain in the general population may also increase. There is a general lack of data regarding the occurrence of chronic pain In ICU survivors, but the few studies that were done have reported prevalences ranging between 33 and 44% (7, 10, 11). The occurrence of newly-acquired pain observed in our study is considerably lower. This is probably caused by the fact that these earlier studies used follow-up periods of 6 months, suffered from imprecision due to small sample sizes (typically around 200 patients), and did not account for bias caused by loss to follow-up (with response rates being only 20-45%).

Several risk factors appeared to be crudely associated with the acquisition of chronic pain after ICU discharge. However, in multivariable analysis only gender and severe inflammation remained. Indeed, a higher risk of pain among females has been previously observed (22), and has been attributed to both biological and psychosocial differences between men and women (25). Likewise, the observed relation between (the duration of) severe inflammation and development of chronic pain in our study confirms previous findings, during which either the presence of sepsis or high CRP values have also been associated with this risk (7, 10). Possibly this is related to specific changes to the somatosensory nervous system that can be triggered by severe inflammation (such as occurs during sepsis), including a reduction of intra-epidermal nerve fiber density (8, 26, 27). As this process has also been linked to neuropathy in these studies, it might also explain the high proportion of patients having features of neuropathic pain observed in our substudy.

Strengths of our study include the relatively large sample size and high response rate. Furthermore, we used multiple imputation techniques to adjust for bias that may have been introduced by non-responders to the survey, and explicitly modeled death as a competing event for the occurrence of pain. However, there are also a number of limitations. First, the survey that was used for pain assessment has not been externally

validated. Although we believe that a customized pain questionnaire offers many advantages over using a better validated, yet non-specific, HRQoL instrument to derive pain information, we did not formally assess its clinimetric performance. Second, it is conceivable that patients already suffering from chronic pain prior to ICU admission may have been misclassified as not having (new) pain, despite significant worsening of their symptoms, as patients themselves judged whether they thought their pain could be related to their ICU admission. Furthermore, in an attempt to make the outcome more robust for information bias that could be caused by such subjective elements, we required a positive response on at least two unrelated questions in order to categorize a patient as having newly-acquired chronic pain. However, together these safeguards may have led to some underestimation of true pain prevalence. Third, we regrettably do not have reliable information on all patients regarding the presence of chronic pain prior to ICU admission. This precludes estimation of overall prevalence of chronic pain in ICU survivors (as opposed to the acquisition of new pain). For the same reason, we also could not assess prior chronic pain as a candidate variable for our prediction model.

Conclusions

Pain is a frequent long-term consequence of critical illness and significantly impacts the daily life of affected patients. Neuropathy appears to be common in patients with chronic pain that could be prospectively evaluated for this, but more studies are necessary to estimate the precise occurrence of various pain features in this population. Although poor predictability limits possibilities for intervention at this time, better knowledge about etiology and risk factors may eventually yield options for prevention and/or preemptive therapy of chronic pain following critical illness.

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SUPPLEMENTARY MATERIAL

Appendix I: Parameterization of predictor variables

Data definitions used for predictor variables that are not self-evident are explained in this appendix.

Neurocognitive deficit	Presence of cerebrovascular disease, depression, and/or dementia
Pressure ulcers	Presence of stage III or IV decubitus, according to the National Pressure Ulcer Advisory Panel (NPUAP) guidelines
Hyperinflammation	Number of days with c-reactive protein (CRP) > 100 mg/L
Organ failure	Number of days with sequential organ failure assessment (SOFA) score > 5
Sedation	Number of days with Richmond agitation sedation scale (RASS) < -2
Delirium	Number of days positive on confusion assessment method for the intensive care unit (CAM-ICU) in patients who were not sedated
Pain	Number of days with numeric rating scale (NRS) for pain ≥ 4 in patients who were not sedated
Opioid use	Days with low, intermediate, and high dose opioids were defined as <30 mg, 30-60 mg, and ≥60 mg oral morphine equivalents per 24 hours, respectively

Appendix II: Distinguishing neuropathic and nociceptive pain characteristics using the DN4

To examine whether patients had pain with neuropathic or nociceptive characteristics, we used the Douleur Neuropathique 4 questions (DN4) during a home visit. This questionnaire consists of seven questions regarding neuropathic characteristics of pain and three neurological examinations of the skin (1). For the physical examination part we used a 10.0 gram monofilament von Frey hair (Touch Test®) to detect pricking hypoesthesia and a soft brush (no. 5, Somedic) to detect touch hypoesthesia and tactile allodynia. The examination was first performed in a non-painful area, preferably on the contralateral side of the painful area, followed by examination of the painful area. Patients experiencing pain in more than one body site were asked to complete the DN4 questionnaire for every painful location separately.

In a systematic review the DN4 and the neuropathic pain questionnaire were regarded as most suitable for clinical use (2). Using a cut-off of 4 or higher for neuropathic pain (range 0-10), a sensitivity and specificity of respectively 74-75% and 76-79% in validation studies in Dutch populations of patients with chronic pain have been reported (3, 4).

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Appendix III: Handling missing data in dataset 1 and 2

Decision rules predictor variables (dataset 1)

For prediction models, it is important that information on predictor variables are easy to obtain in several settings. Therefore, we chose to develop pragmatic decision rules to replace missing values in predictor variables. For in total 27 predictors and 13387 observation days, we had only missing data in the following daily measured variables: CRP 3%, SOFA <1%, sedation <1%, delirium 4%, and pain 5%. Also, there were <1% missing values for pressure ulcers. Used decision rules are: 1) the last day of ICU admission was only taken into account when a patient was discharged or transferred after 6AM; 2) the value of the day before the missing observation was carried forward; and 3) the value of the day after the missing observation was carried backward.

After applying these rules in this order there were still 98 missing observations for CRP, 6 for sedation, 247 for pain, 75 for delirium, and 6 for pressure ulcers (all <2% of the total amount of observation days). These remaining missing values were replaced with the median (for CRP) or most frequent (for sedation, delirium, and pressure ulcers) observed value for that particular day of ICU admission. For pain there was a slightly different rule: it was replaced with the most frequent observed value for that particular day of ICU stay unless the patient received a high doses of opioids the same day.

Multiple imputation procedures for missing outcomes (dataset 1) and pain characteristics (dataset 2)

We used multiple imputation procedures to avoid bias by selective non-response and assumed that data were missing at random (1,2). For these procedures, we used an imputation method based on chained equations (R-package 'mice', version 3.4.0, 2019) (3, 4). We used demographic, clinical and follow-up data in our imputation models to replace missing values. The stability of imputations was evaluated by checking summary statistics and density plots for each imputed variable (4-7).

There were 410 patients (30%) with missing outcome information among the 1368 one-year survivors in dataset 1. Considering this non-response of 30%, we performed 30 imputations with 30 iterations per imputation. Among the 160 responders with newly-acquired chronic pain, the amount of missing values differed strongly per question: average pain intensity had 12% missing values, bodily locations <1% missing, impact on daily life 11-13% missing values per domain, and health-related quality of life (HRQoL) <1% per domain. Considering that the highest amount of missing values was 13%, we performed 10 imputations with 25 iterations per imputation.

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Appendix IV: Table S.1. Demographics and characteristics of ICU stay in patients who died during follow-up and who survived with or without newly-acquired chronic pain (n=1842)

	Patients who died during follow-up	Patients with or without newly-acquired chronic pain	
	n=474	n=1368	P-value
Age, years	68 (59-75)	59 (46-69)	<0.001
Gender, male	304 (64.1)	880 (64.3)	0.984
Comorbidities			
- Diabetes mellitus	93 (19.6)	190 (13.9)	0.004
- Any malignancy	146 (30.8)	212 (15.5)	<0.001
- Neurocognitive deficit[a]	110 (23.2)	207 (15.1)	<0.001
- Connective tissue disease	19 (4.0)	48 (3.5)	0.720
- Obesity	99 (20.9)	295 (21.6)	0.806
Drug use before ICU admission			
- Opioids	56 (11.8)	90 (6.6)	<0.001
- Antineuropathic drugs	19 (4.0)	34 (2.5)	0.121
APACHE IV score	84 (69-104)	7 (55-86)	<0.001
Admission type			<0.001
- Medical	270 (57.0)	667 (48.8)	
- Surgical elective	75 (15.8)	330 (24.1)	
- Surgical non-elective	129 (27.2)	371 (27.1)	
Surgical procedure during ICU stay	127 (26.8)	441 (32.2)	0.031
Pressure ulcers[b]	125 (26.4)	237 (17.3)	<0.001
Number of days with			
- Hyperinflammation (CRP > 100 mg/L)	3 (1-5)	2 (1-5)	0.899
- Organ failure (SOFA > 5)	2 (1-5)	2 (0-4)	<0.001
- Mechanical ventilation	5 (2-10)	4 (2-8)	0.013
- Sedation (RASS < -2)	2 (0-5)	1 (0-4)	0.027
- Deliriumc	0 (0-2)	0 (0-2)	0.307
- Pain (NRS > 3)d	0 (0-1)	1 (0-2)	<0.001
- Opioid use[e]			
- Low dose	3 (1-6)	2 (1-4)	<0.001
- intermediate dose	2 (1-3)	2 (1-3)	0.582
- High dose	0 (0-2)	1 (0-3)	<0.001
Other analgesic drug use			
- Paracetamol/NSAID	446 (94.1)	1326 (96.9)	0.008
- Antineuropathic drugs	39 (8.2)	135 (9.9)	0.336
- Alpha-2 agonists	188 (39.7)	654 (47.8)	0.003
- Epidural or locoregional analgesia	26 (5.5)	90 (6.6)	0.462
ICU length of stay, days	6 (4-13)	6 (3-11)	0.164

APACHE: acute physiology and chronic health evaluation, ICU: intensive care unit, CRP: C-reactive protein, SOFA: sequential organ failure assessment, RASS: Richmond agitation-sedation scale, NRS: numeric rating scale, NSAID: nonsteroidal anti-inflammatory drugs. Continuous and categorical data are presented as medians (first – third quartile) and frequency (percentage), respectively. [a] Cerebrovascular disease, depression, and/or dementia. [b] Stage III or IV, according to the National Pressure Ulcer Advisory Panel (NPUAP) guidelines. [c] Daily assessed using the confusion assessment method for the intensive care unit (CAM-ICU) in patients who were not sedated. [d] Daily assessed using a numeric rating scale in patients who were not sedated. [e] Days with low, intermediate and high dose opioids were defined as <30, 30-60, and ≥60 mg oral morphine equivalents per 24 hours, respectively.

Appendix V: Table S.2. Demographics and characteristics of ICU stay in patients included in the substudy (n=42)

	Patients with neuropathic pain	Patients without neuropathic pain	
	n=21	n=21	P-value
Age	55 (44-60)	54 (48-65)	0.725
Gender, male	13 (62)	9 (43)	0.354
Comorbidities			
- Diabetes Mellitus	2 (10)	1 (5)	1.000
- Any malignancy	1 (5)	1 (5)	1.000
- Neurocognitive deficit	0 (0)	3 (14)	0.232
- Connective tissue disease	1 (5)	0 (0)	1.000
- Obesity	6 (29)	3 (14)	0.454
Pain prior to ICU admission[a]	10 (48)	5 (24)	0.710
Drug use before ICU admission			
- Opioids	2 (10)	0 (0)	0.488
- Antineuropathic drugs	1 (5)	0 (0)	1.000
APACHE IV Score	72 (47-81)	69 (59-93)	0.513
Admission type			0.806
- Medical	12 (57)	10 (48)	
- Surgical elective	2 (10)	2 (10)	
- Surgical emergency	7 (33)	9 (43)	
ICU length of stay	7 (6-19)	7 (4-9)	0.181

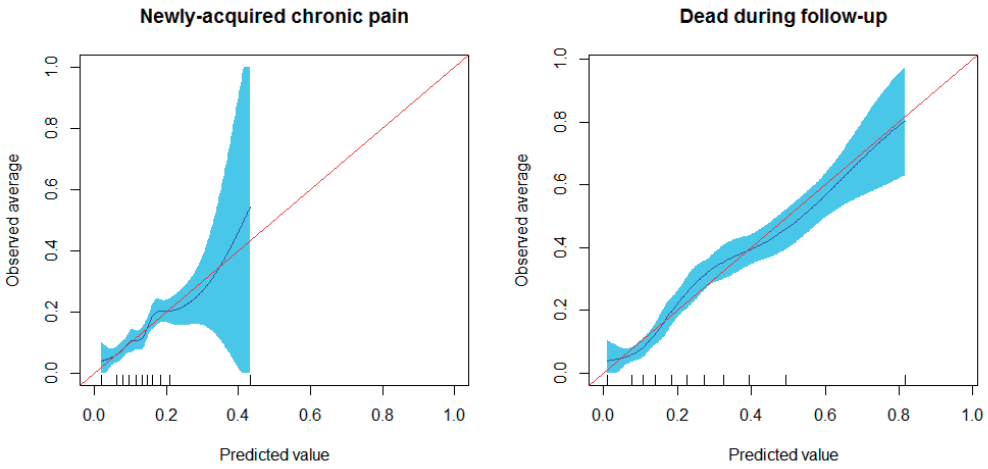
APACHE : acute physiology and chronic health evaluation, ICU: intensive care unit. Continuous and categorical data are presented as medians (first – third quartile) and frequency (percentage), respectively. Neurocognitive deficit: cerebrovascular disease, depression, and/or dementia. [a] Respectively 6 and 7 missing values.

Appendix VI: Table S.3. Predictor variables for death during follow-up

	Coefficient	OR (95% CI)	P-value
Intercept	-3.092	-	<0.001
Age, years[a]	0.009	1.01 (0.98-1.04)	0.555
- Age, years'	0.032	1.03 (0.98-1.09)	0.226
- Age, years''	-0.131	0.88 (0.60-1.28)	0.496
Gender, male	-0.080	0.92 (0.72-1.18)	0.524
Any malignancy	0.728	2.07 (1.58-2.71)	<0.001
APACHE IV score	0.012	1.01 (1.01-1.02)	<0.001
Days with hyperinflammation[b]	-0.012	0.99 (0.93-1.03)	0.581
Days with sedation[c]	0.100	1.10 (1.06-1.15)	<0.01
Days with pain[d]	-0.081	0.92 (0.85-1.01)	0.068
Days with high dose opioids[e]	-0.102	0.90 (0.84-0.96)	0.003
ICU length of stay, days	0.004	1.00 (0.99-1.01)	0.428

Regression coefficients were estimated using a multivariable multinomial logistic regression model that included death, surviving with newly-acquired chronic pain, and surviving without pain one year after ICU discharge as discrete outcome categories. Predictor selection was based on the least absolute selection and shrinkage operator (LASSO). Only coefficients for newly acquired chronic pain after critical illness are shown here; coefficients for death during follow-up are reported in Table S.3. APACHE: acute physiology and chronic health evaluation, ICU: intensive care unit. For all 'days with' the first 14 days of ICU admission are taken into account. [a] Modelled as a restricted cubic spline with 4 knots, namely 25, 55, 67, and 80 years. [b] C-reactive protein > 100 mg/L, [c] Richmond agitation sedation-scale < -2, [d] Numeric rating scale > 3, [e] ≥ 60 mg oral morphine equivalents per 24 hours.

Appendix VII: Figure S.1. Calibration plots for newly-acquired chronic pain and death during follow-up with 95% confidence intervals



7

Chapter 7

Non-proportionality in Cox regression models and its implications for the interpretation of survival data in critically ill patients

Maria E. Koster-Brouwer
Marinus J.C. Eijkemans
Olaf L. Cremer

In submission

Abstract

Introduction Although proportional hazards (PH) underpin Cox regression models, alertness on possible violation of this assumption is frequently lacking in reports of clinical research in critically ill patients. We demonstrate two mechanisms that can lead to non-proportionality, and assess their impact on effect estimates in survival analysis.

Methods To illustrate bias caused by confounding, we used clinical data and assessed the association between early acute kidney injury (eAKI) and mortality in critically ill patients with sepsis. To illustrate bias caused by unobserved heterogeneity, we generated survival data and simulated two hypothetical effects representing this phenomenon. Subsequently, we demonstrate several methods to graphically check the PH assumption. Furthermore, we statistically assessed non-proportionality by the weighted residuals score test. In case of non-proportional hazards, we added time-interactions to allow the HR to change over time.

Results Among 1418 patients in the clinical dataset, 520 (37%) developed eAKI. Both univariable and multivariable Cox regression suggested that there was a significant association with mortality (crude hazard ratio (HR) 1.63 (95%CI 1.40-1.89; p<0.001), adjusted HR 1.26 (95%CI 1.08-1.48; p=0.004), respectively). However, neither analysis met the PH assumption as evidenced by graphical checks and the weighted residuals score test (p<0.001), although non-proportionality was reduced somewhat by adding covariates to the model. In fact, negative interaction with time since sepsis onset suggested that the effect of eAKI on mortality was present only during the first weeks after its occurrence, but not thereafter. Using simulated data, we then demonstrated that remaining heterogeneity in the hazards could be caused by an unmeasured risk factor for mortality not associated with eAKI (i.e., unobserved heterogeneity). Extending a univariable model that violated the PH assumption with a simulated risk factor for mortality removed non-proportionality and restored the HR to its simulated value.

Conclusions Both confounders and risk factors for the study outcome can cause non-proportional hazards due to (unobserved) heterogeneity in the study population. Verifying the PH assumption should therefore be a systematic part of doing research using Cox regression models.

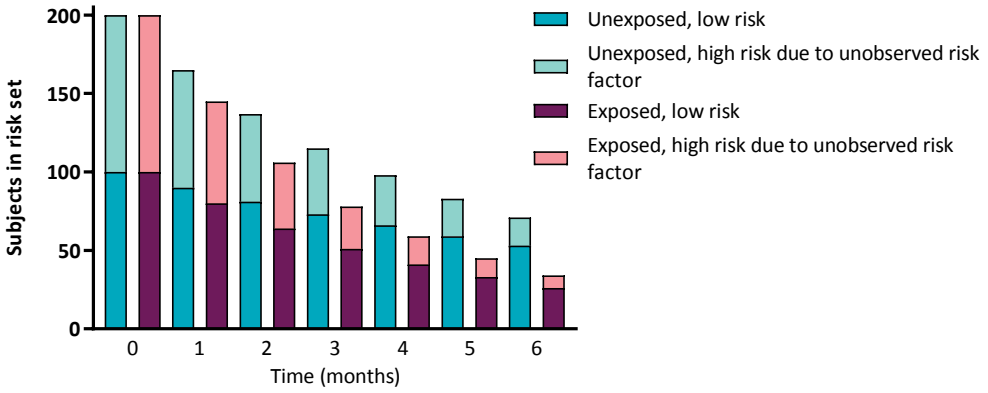
Introduction

When assessing patient-centered outcomes in intensive care unit (ICU) patients, studies increasingly focus on mortality and morbidity beyond hospital discharge. These outcomes are commonly analyzed by Cox proportional hazards (PH) models using time-to-event data. The effect estimates generated by such models are represented as a hazard ratio (HR), which can be defined as the ratio between the event rate (of a particular outcome of interest) in the exposed and unexposed group for each (arbitrary) moment during study follow-up. Hence, meaningful interpretation of HRs requires a constant relation between these event rates over time, i.e. the PH assumption (1, 2).

Unfortunately, violation of the PH assumption is not uncommon. Basically, it can arise from either one of two conditions. The first is observed when the impact of a variable in the model varies over time, for instance when the impact of a determinant is large in the first days or weeks after intensive care unit (ICU) admission, but no difference in outcome rates is observed between exposed and non-exposed subjects during later time-periods. It is important to notice that, for the PH assumption to hold, not only the determinant of interest but all covariates in the model must satisfy this condition (3). The second mechanism can be observed when an important risk factor for the outcome under study is not included as a covariate in the analysis (1). This is referred to as unobserved heterogeneity due to an omitted variable (4). Even if this omitted variable is equally distributed between the exposed and non-exposed groups (i.e., it does not meet the definition of a confounder), this variable can disturb the exposure-outcome association under study. This is due to a specific situation that arises in time-to-event data, where the risk set changes continuously. Subjects who are prone for the outcome under study disappear more quickly from the risk set than those with a better prognosis (4). Due to this 'depletion' of the risk set, the risk factor starts to behave like a confounding variable as time elapses (i.e., the even distribution between exposure groups becomes distorted). This process is graphically depicted in Figure 1.

As short and long-term mortality often do not share the same risk factors, and considering the complexity of causes for long-term mortality in critically ill patients, it has been suggested that violation of the PH assumption might occur regularly when analyzing survival data in critically ill patients (5-7). Still, when Cox regression is used model assumptions frequently remain untested and underreported (8-10). This suggests that the phenomenon is generally poorly understood, and its implications for interpretation of study results underestimated. For these reasons, we aimed to illustrate the occurrence of non-proportionality by the aforementioned mechanisms and demonstrate its consequences for studying survival data in ICU patients.

Figure 1. Graphical display of the influence of an omitted risk factor on risk set composition



This figure depicts the situation in which an unobserved risk factor for the outcome under study – not associated with the exposure variable – is present in the data. Subjects prone for the outcome under study (i.e., exposed) disappear more quickly from the risk set than those with a better prognosis. Due to this phenomenon, the unobserved risk factor starts to behave like a confounder over time and can cause non-proportional hazards.

Methods

We analyzed the association between early acute kidney injury (eAKI) and case fatality occurring up to one year after ICU admission using univariable and multivariable Cox regression analyses in both clinical and simulated data.

Clinical data

To illustrate the occurrence of non-proportionality and its potential consequences in general research data, we analyzed the association between early acute kidney injury (eAKI) and mortality in a cohort of patients admitted to the ICU of the University Medical Center Utrecht or the Academic Medical Centrum in Amsterdam with sepsis between January 2011 and May 2015. All included patients met sepsis-3 criteria upon presentation to the ICU (i.e., had organ failure due to clinically suspected infection as defined by criteria published previously (11)) and a length of stay of at least 24 hours. Patients with chronic renal failure (requiring dialysis) and patients who had been transferred from another ICU were excluded. Off note, this study was not designed to demonstrate existence of an (etiologic) association between eAKI and mortality in critically ill patients.

Renal function was assessed using the RIFLE criteria (12). Early AKI was defined as the presence of RIFLE class 'injury' or 'failure' on the first day in ICU. The primary end point for the analysis was time to death, which was queried from the Dutch municipal population registers. Confounders accounted for in the analyses were based on

previous research by our group (13) and included age, gender, pre-ICU hospital length of stay, admission type (i.e. medical, surgical elective, or surgical emergency), Charlson comorbidity index (14), the need for mechanical ventilation, and the acute physiology component (APS) of the APACHE IV score without the creatinine component (15).

Simulated data

To illustrate non-proportionality by unobserved heterogeneity, we simulated one-year survival data for 5000 subjects using R-package simsurv (16). Time to death was randomly drawn from a Weibull distribution using a λ of 0.009 and a γ of 0.55 to generate skewed time-to-event data. Simulated early AKI (eAKI_sim) data were generated to represent a prevalence of approximately 30%, and an association with mortality corresponding to a HR of 2.9. A hypothetical (unobserved) risk factor for death was additionally simulated (Z_sim), having a prevalence of approximately 50% and a HR of 4.1 for mortality being independent of eAKI_sim. This risk factor was subsequently used to study the effect of an omitted covariable on the estimated hazard function. Of note, we carefully ensured that the HRs for mortality associated with eAKI_sim as well as Z_sim would remain constant over time in order not to violate the PH assumption. We deliberately simulated data having strong associations with mortality for eAKI_sim and Z_sim, to be able to clearly demonstrate the effect of an unobserved risk factor on proportionality of the hazards and the estimated effect. In practice, unobserved heterogeneity could also be caused by co-occurring risk factors which individually have smaller effects on the outcome under study. The simulated dataset was replicated 50 times and the analysis was performed on each dataset in order to check for consistency of results.

Survival analysis

We constructed univariable and multivariable Cox regression models. For continuous covariates we first evaluated what the best fitting functional form (e.g., linear, logarithmic, spine) was before incorporation it in the multivariate model. To graphically inspect the proportionality of the hazards we used three common methods, as appropriate. First, Log-Log survival probability plots were made and visually inspected (17, 18). Second, we assessed cumulative Martingale residuals. This method simulates various paths of the hazard function over time under the PH assumption and plots these paths together with observed data (19). Third, we plotted scaled Schoenfeld residuals and evaluated their (in)dependence on time based on the slope of the residuals. Of note, for examination of the PH assumption in the multivariable Cox regression analysis only the scaled Schoenfeld residuals were used, as the others methods are not suited to use when evaluating multivariable models (20, 21). The occurrence of non-proportionality was statistically tested using the weighted residuals score test based on the Schoenfeld residuals.

As a last step, interactions between each covariate and the logarithmic function of time were added to the multivariable model to create time-dependent HRs. A significant interaction with time was interpreted as a change of the HR over time, i.e., non-proportional hazards. These significant interactions were incorporated in the model whilst non-significant were removed for the sake of simplicity.

Continuous variables are presented as medians with interquartile quartile, dichotomous and categorical variables as absolute numbers with percentages. Differences between groups were statistically tested using the X^2 test or Mann-Whitney U test, as appropriate. All analyses were performed in R studio (R Studio Team 2015, Boston, MA).

Results

Data descriptives

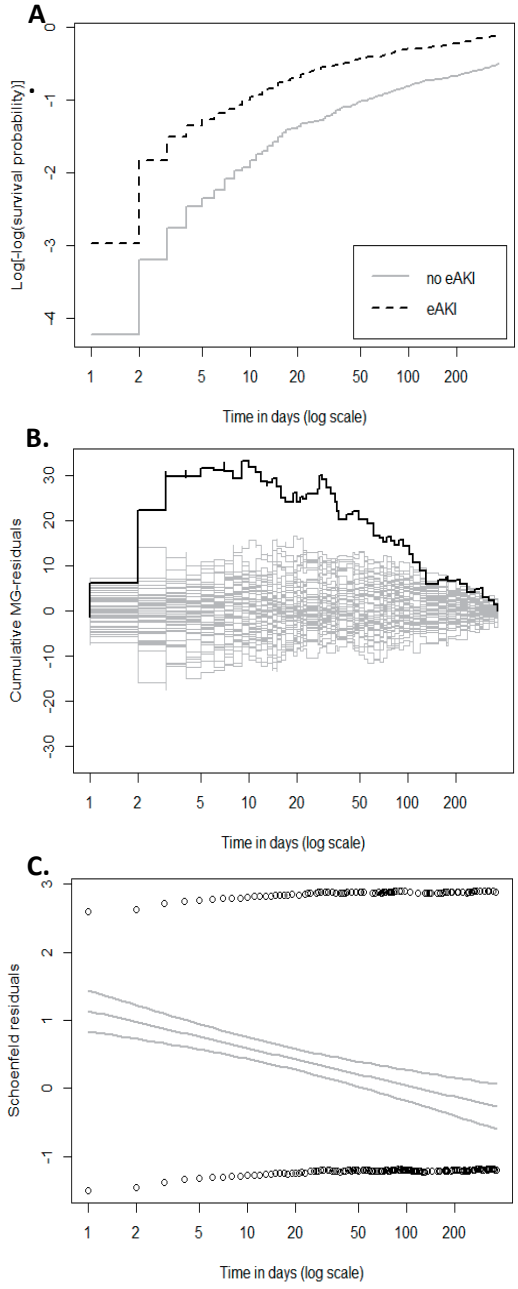
Among 1545 subjects in the clinical dataset who had been admitted with sepsis to the ICU, 1418 fulfilled eligibility criteria and were included in the analysis (Appendix I: Figure S1). In this population, eAKI occurred in 520 (37%) patients (Appendix II: Table S1). Observed case fatality rates in ICU, in hospital, and after one year were 40%, 49%, and 59% versus 18%, 30%, and 45% for patients with and without eAKI, respectively.
In the simulated dataset, the occurrence of eAKI was randomly assigned to 1525 (30.5%) of 5000 virtual subjects. Mortality rates during the simulated time period of one year in subjects with and without eAKI were 1098 (72.0%) and 1428 (41.1%), respectively. The unobserved risk factor for mortality (represented by the omitted variable, Z_sim) was allocated to 51.4% and 50.1% of subjects with and without eAKI.

Violation of the PH assumption by time-dependent impact of the exposure

Based on univariable Cox regression analysis, the crude HR for mortality (i.e., death across the 1-year follow-up period) was estimated to be 1.63 (95%CI 1.40-1.89, $p<0.001$) when comparing patients with and without eAKI. We then investigated the PH assumption in three graphical ways (Figure 2).

In case of true proportionality of the hazards, the log-log survival probability plots of exposed and unexposed subjects should remain parallel over time. However, non-proportionality was indicated by the convergence of these survival curves during follow-up (Figure 2A). Second, cumulative Martingale residuals were used plotting standardized score processes that would have been observed when the model was correctly specified (i.e., having proportional hazards) and compared with the observed standardized score process. The observed path should be typical of the simulated paths in case of proportionality of the hazards, which was not the case as the black (observed) path clearly deviates from the grey (simulated) paths indicating non-proportionality

Figure 2. Graphical checks of the proportional hazards assumption for the univariable association between early acute kidney injury and one-year mortality in patients with sepsis (n=1418)



eAKI: early acute kidney injury. Part A represents log-log survival probability plots, part B cumulative martingale residuals, and part C Schoenfeld residuals. All parts indicate non-proportionality of the hazards.

(Figure 2B). Third, the Schoenfeld residuals represent the correlation between the residuals of a variable of interest and time. Independency of time, as evidenced by a random pattern of the residuals against time, will be observed in case of proportional hazards. As independence of time was difficult to evaluate because eAKI is a categorical variable resulting in residuals that were not scattered, we fitted a smooth curve and confidence interval across the residuals. In case of PH, it should be possible to draw a straight, horizontal line between the upper and lower limit of the confidence interval. This was not the case, further suggesting the presence of non-proportional hazards (Figure 2C). As a final step, the weighted residuals score test based on the Schoenfeld residuals confirmed that the PH assumption was violated in this univariable model (X^2 test-statistic 26.3, $p<0.001$).

Violation of the PH assumption by confounding

The crude HR for mortality decreased after inclusion of potential confounders into the Cox regression model (adjusted HR 1.26, 95%CI 1.08-1.48). To assess the PH assumption of this multivariable model, we plotted scaled Schoenfeld residuals as explained previously, but this time for each included covariate separately (Appendix III: Figure S2). This, again, suggested violation of the PH assumption, which was confirmed by the weighted residuals score test (X^2 test-statistic 7.66, $p<0.001$). However, the observed decrease in this test-statistic suggests that at least part of non-proportionality in the univariable model was caused by confounding variables for which this multivariable model could account. To allow the model to estimate HRs that changed over time for variables causing non-proportionality, we included interaction terms between eAKI, gender, age, Charlson comorbidity index, pre-ICU hospital length of stay, and APS score (Table 1). Based on this model, we observed an HR for eAKI that started at 1.98 but quickly decreased to a value not statistically different from 1 as indicated by the 95% confidence intervals (Figure 3).

Violation of the PH assumption by unobserved heterogeneity

We performed further analyses of non-proportionality in a simulated dataset with a predetermined association between eAKI_sim and mortality, and a hypothetical unobserved risk factor Z_sim for death that had no relation with this exposure (i.e., was not a confounder). Despite the fact that these data were purposefully generated to represent a hazard function that was perfectly constant over time, univariable Cox-regression analysis in this dataset resulted in a crude HR estimate of 2.46 (95%CI 2.27-2.66, $p<0.001$) rather than its 'true' value of 2.9. Furthermore, both graphical inspection of the residuals and the weighted residuals score test appeared to indicate that the PH assumption was violated (weighted residuals score test-statistic 4.54; $p=0.033$). However, after inclusion of the omitted variable Z_sim into the regression model, proportionality of the hazards over time was completely restored (weighted

Table 1. Multivariable Cox regression model including (significant) interactions between covariates and the logarithmic function of time

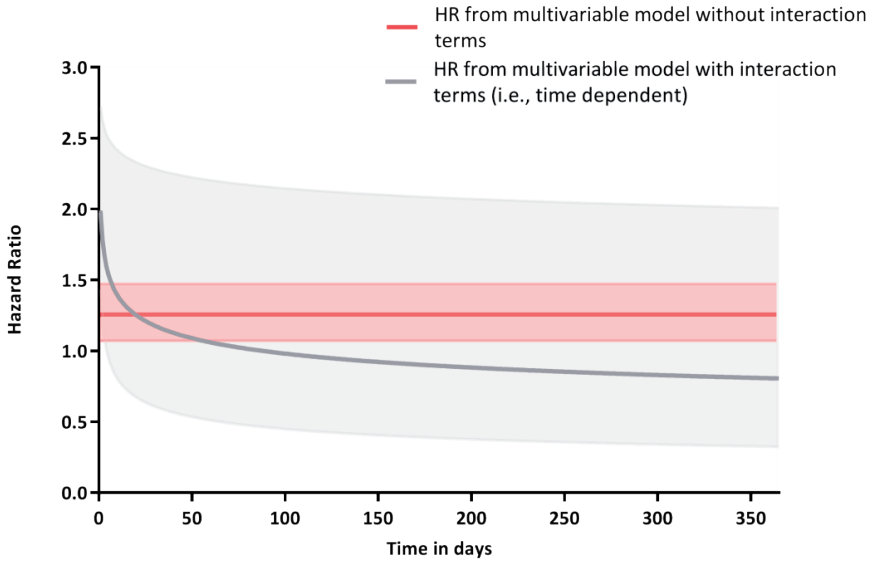
	Coefficient	Hazard ratio	95% CI	P-value
No eAKI *(reference group)*	-	-	-	-
eAKI	0.68	1.98	1.44-2.72	<0.001
Gender, male	-0.20	0.82	0.61-1.10	0.189
Age	0.01	1.01	1.00-1.02	0.235
Admission type: surgical elective	-0.20	0.82	0.62-1.09	0.169
Admission type: surgical emergency	0.04	1.04	0.85-1.27	0.702
Charlson comorbidity index	0.00	1.00	0.93-1.09	0.921
Chronical renal insufficiency	0.08	1.08	0.86-1.36	0.488
Pre-ICU hospital length of stay	-0.11	0.90	0.81-0.99	0.034
APS without creatinine component (quadratic)	0.0001	1.0001	1.0001-1.0002	<0.001
Septic shock at admission	0.18	1.20	1.02-1.42	0.025
Mechanical ventilation at admission	-0.10	0.91	0.72-1.15	0.427
eAKI * log(time)	-0.15	0.86	0.78-0.95	0.003
Gender, male * log(time)	0.10	1.10	1.00-1.21	0.041
Age * log(time)	0.00	1.00	1.00-1.01	0.015
Charlson comorbidity index * log(time)	0.04	1.04	1.01-1.06	0.003
Pre-ICU hospital length of stay (log) * log(time)	0.06	1.06	1.03-1.10	<0.001
APS without creatinine component (quadratic) * log(time)	-0.0000	1.0000	1.0000-1.0000	<0.001

CI: confidence interval. eAKI: early acute kidney injury. ICU: intensive care unit. APS: acute physiology score.

residuals score test-statistic 0.03; p=0.858). Furthermore, the model now estimated a HR of 2.83 (95%CI 2.61-3.06) for the exposure-outcome relation, which was very similar to the 2.9-fold effect simulated.

In 9 out of 50 dataset replications, the weighted residuals score-test statistic did not indicate violation of the PH assumption when not taking into account Z_sim, with p-values ranging between 0.0559 and 0.782. However, when repeating the procedure using a rank instead of natural log transformation of the time scale, in 97% of the cases the test performed according to what was expected based on the simulated data.

Figure 3. Estimated hazard ratios and 95% confidence intervals from multivariable models with and without interactions between covariates and the logarithmic function of time



HR: hazard ratio.

Discussion

In this study, we demonstrated methods to check for the presence of non-proportional hazards in clinical survival data, and provide meaningful interpretation to the HR in such a situation. In our case study example, the estimated HRs for eAKI based on the univariable and multivariable model were distorted due to violation of the PH assumption. Naïve interpretation of these HRs would have led to the conclusion that during the first year after ICU admission the risk of dying is higher for patients who were admitted with or developed AKI in the first day of ICU admission. In fact, this statement holds only true for a short period of time after ICU admission; the HR underestimated the effect in the first weeks after eAKI and overestimated the effect for the remaining duration of follow-up. Multivariable adjustment for potential confounders reduced non-proportionality, but significant heterogeneity remained. Using simulated data in addition to our clinical data, we studied a 'hidden' mechanism that may lead to non-proportional hazards. We demonstrated that remaining heterogeneity after multivariable adjustment could have been caused by a risk factor for mortality not taken into account in the Cox regression model. This unobserved heterogeneity could influence the effect estimate, and might cause non-proportionality whilst in reality the hazards are constant over time.

In studies using survival data from critically ill patients, for example patients with sepsis, non-proportionality might occur frequently. This population is extremely heterogeneous and differs strongly with respect to age, comorbid status, and genetic profiles (22). This type of heterogeneity has been observed in sepsis trials and is reflected in mortality rates that differ strongly per study which can only partially be explained by (reported) patient characteristics (6). Some authors therefore even have proposed that the PH assumption probably does not hold in sepsis as many factors often present in critically ill patients with sepsis are known to pose different hazards of death over time (5). Baring this in mind, one could say beforehand that it would be challenging to meet the PH assumption when analyzing survival data in such a complex study population.

The objective of our study was to create awareness of the PH assumption when modelling time-to-event data using Cox regression analysis by clinical researchers and physicians. Verifying proportionality of the hazards should be a systematic procedure in the analysis of time-to-event data with Cox regression models. When using Cox regression models, considerable thought should be given to variables possibly confounding the studied association, but one should be aware that risk factors for the study outcome – not associated with the study determinant – can also influence the estimated effect. Extension of the model with such variables might reduce underlying heterogeneity in the study population. Another argument to aim for PH is that non-proportionality does not only have consequences for obtained effect estimates, but that it can also lead to inferior model fit and suboptimal power (23). Special caution is advised in situations with a long follow-up period, in heterogeneous populations, and in studies with small sample sizes as the latter influences the power of statistical tests to detect non-proportionality.

Conclusions

Consideration of the proportionality of hazards is crucial when interpreting Cox regression models as violation of this assumption may lead to distorted estimates of the HR, only representing an average effect and not the ratio in hazards per time unit. This might lead to erroneous conclusions regarding the effect under study. A thorough examination of the proportionality of the exposure-outcome relation using graphical methods and formal statistical tests is therefore encouraged. The incorporation of confounders as well as risk factors for the study outcome might be a solution for heterogeneity of the hazards in the study population.

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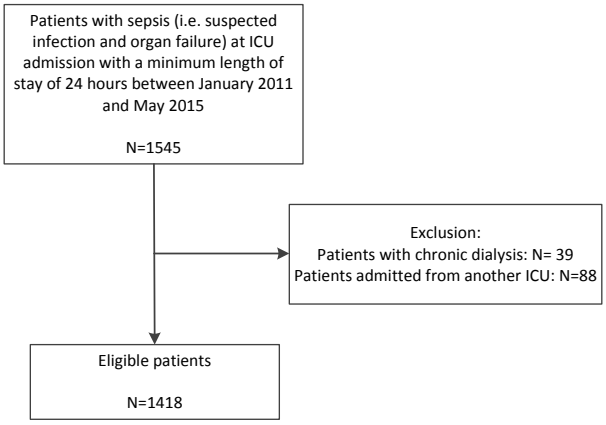
SUPPLEMENTARY MATERIAL

Appendix I: Figure S1. Flowchart of patient inclusion



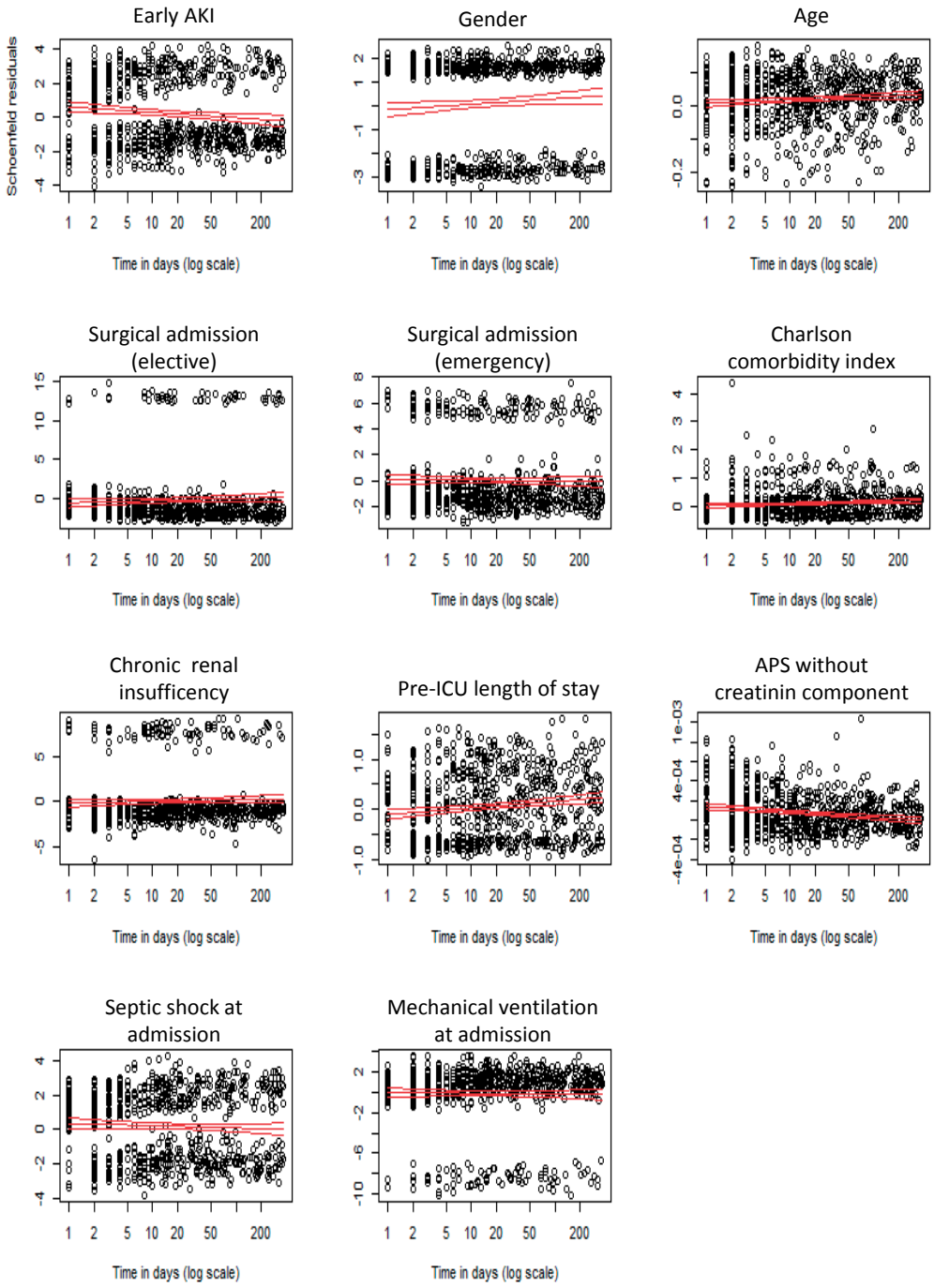
ICU: intensive care unit.

Appendix II: Table S1. Patient characteristics (n=1418)

	No eAKI (n=898)	eAKI (n=520)	p-value
Gender, male	562 (63)	314 (60)	0.445
Age, years	62 (51-72)	65.5 (55-73)	<0.001
Admission type			0.512
- **Medical**	658 (73)	376 (72)	
- **Surgical elective**	81 (9)	41 (8)	
- **Surgical emergency**	159 (18)	103 (20)	
Charlson comorbidity index	1 (0-2)	1 (0-2)	0.042
Chronic renal insufficiency	76 (9)	80 (15)	<0.001
Pre-ICU hospital length of stay, days	1 (0-7)	1 (0-7)	0.881
APACHE IV score	80 (64-97)	100 (79-124)	<0.001
Septic shock at admission	358 (40)	337 (65)	<0.001
Mechanical ventilation at admission	793 (88)	445 (86)	0.160
ICU length of stay, days	6.1 (3.1-11.7)	6.0 (2.6-13.9)	0.420

eAKI: early acute kidney injury. ICU: intensive care unit. APACHE: acute physiology and chronic health evaluation. Continuous variables are shown as medians (1^{st} quartile – 3^{rd} quartile), dichotomous/categorical variables are shown as n (%). Differences between groups were statistically tested using the X^2 test or Mann-Whitney U test, as appropriate.

Appendix III: Figure S2. Scaled Schoenfeld residuals for all variables in the multivariable model



AKI: acute kidney injury. ICU: intensive care unit. APS: acute physiology score.

8

Chapter 8

Summary & General discussion

The aim of this thesis was to asses the societal impact of sepsis and inflammation in critically ill patients, both during and following ICU admission. We investigated this by exploring whether a novel diagnostic test could be of clinical benefit (Part I), to estimate the potential economic value of this new test we estimated (a) costs associated with sepsis treatment in the ICU, in particular expenditure directly related to the diagnostic process, and (b) costs associated with long-term survivorship (Part II), and to understand what drives these long-term costs we studied specific late sequelae occurring in ICU survivors of sepsis (Part III).

PART I. DIAGNOSING INFECTION

Sepsis is associated with high mortality as well as long-term morbidity. Rapid intervention may alleviate some of the disease burden, yet is hampered by the fact that early detection and accurate diagnosis of sepsis are difficult. In recent years, many biomarkers involved in the host-response have been proposed to diagnose infection, for example CRP, procalcitonin, and interleukin-6 (1-3). Nevertheless, several studies indicated that single biomarkers have only limited accuracy in diagnosis of infection (2, 4-7), and research started to focus on more complex biomarker panels (1, 8). One of those is the SeptiCyte LAB™ test (Immunexpress, Seattle, WA), a host-response assay based on quantitative reverse transcriptase PCR methods. This test has been reported to be able to distinguish infection from sterile inflammation during critical illness with high discriminative power (9, 10). However, these studies had a rather explanatory approach using highly selected study populations, precluding firm conclusions about clinical utility.

In **Chapter 2** we performed a clinical validation study investigating the diagnostic and prognostic performance of the SeptiCyte LAB test in a cohort of 467 hospitalized patients admitted to the ICU with acute respiratory failure. The reference standard was a prospective adjudication of the presence of infection at ICU admission using all patient, clinical, radiological, and microbiological data that became available during ICU stay. Daily, these data were carefully reviewed by trained researchers to categorize the likelihood of infection as none, possible, or confirmed. Using the subjects in whom infection was either ruled-out or confirmed, we found that the sensitivity of the test was good (96%), but the amount of false-positive results was high, resulting in a specificity of 18%. Diagnostic performance of SeptiCyte LAB was moderate (AUC 0.727; 95%CI 0.666-0.788) and was not superior (p=0.919) to the easily available and less costly biomarker CRP (AUC 0.731; 95%CI 0.677-0.786). Although there are no market prices of SeptiCyte LAB available yet, it is expected that it will be far more expensive than measuring CRP, in many hospitals being a routine laboratory test. Furthermore, prognostication did not improve when using the SeptiCyte LAB results in combination with the APACHE IV score, a commonly used severity of illness index.

Considerations

Our study on the diagnostic performance of SeptiCyte LAB showed less favourable results than other studies evaluating this test (9, 10). An important explanation for this difference might be the approach that we used. In diagnostic research, two approaches can be distinguished, namely the explanatory and pragmatic approach. The main aim of an explanatory study design is to evaluate the diagnostic performance of a test in a controlled setting (e.g. highly selected patients, smaller samples), whilst the aim of a pragmatic design is to reflect the clinical conditions in which a test is supposed to be used (e.g., larger cohorts of consecutive patients within a certain domain) (11, 12). In **Chapter 2** we used the latter, in contrast to other studies reporting on SeptiCyte LAB using a more explanatory approach, which likely resulted in an overestimation of true diagnostic performance. Pragmatic studies in diagnostic research are necessary to provide insight in the utility of a test when introduced in clinical practice. It is undesirable that a novel and costly test for such a harmful disease as sepsis is introduced in clinical care based on research not reflecting daily clinical practice, and thus not reflecting true clinical utility.

The pragmatic approach also led to inclusion of patients in whom infection was not evidently absent or present, but that infection could also be part of the differential diagnosis. The choice of study population was the topic of author correspondence with the developers of the test (included in the **addendum to chapter 2**). McHugh et al. argued that in our patient population the risk of diagnostic misclassification by the reference test was higher than in previous studies, and that this could have led to underestimation of the performance of SeptiCyte LAB. In our letter we argue why we feel that our study results are an accurate reflection of diagnostic performance and that it is of greatest importance to study diagnostic performance in a cohort mimicking a population with a clinical need for such a test. For example, if assessing the cost-effectiveness of a diagnostic test it could only be reliably determined in such pragmatic studies.

The occurrence of diagnostic misclassification in our study and other studies cannot be estimated as there is no gold standard for infection. Yet, in our study we were transparent about the difficulty of diagnosing infection by not only reporting test results in patients in whom infection could be ruled-out or confirmed, but also in patients in whom infection was – though using all available patient, clinical, radiological, and microbiological data – still undetermined. In calculations of diagnostic performance measures it was not possible to include these latter patients as dichotomization of the infection diagnosis was required. Hence, in contrast with what the developers stated, these calculations are probably overestimating the true clinical value of the test. Thus, although we cannot rule-out diagnostic misclassification, it is unlikely that it explains the observed difference in diagnostic performance between our study and previous reports. Indeed, our cohort consisted partly of patients in which presence or absence

of infection was difficult to establish upon presentation to the ICU, but this is exactly the population in whom there is the highest need for such a test and in whom the largest effect in terms of fastening diagnostic processes can be achieved.

Future perspectives and implications

- To establish the diagnostic performance and robustness of diagnostic tests, pragmatic studies in large patient cohorts in different settings are needed.
- The only way to estimate real clinical utility of a diagnostic test for infection is an intervention trial, not only evaluating diagnostic performance, but also its impact on patient outcomes and health care expenditures.

PART II. ESTIMATING ECONOMIC BURDEN

Sepsis is associated with high severity of disease and prolonged ICU length of stay, but even beyond the acute phase of critical illness patients may have problems with their physical and mental health (13). Both the time spent in ICU and beyond discharge might therefore cause substantial health care expenditure (14-16). Previous studies investigating sepsis-associated health care costs have been hampered by the use of administrative data to identify sepsis and estimate expenditure, which inhibits the calculation of health care costs on a patient level. Other studies have used a limited time-horizon over which costs were assessed, prohibiting the adjustment for trends in health care costs, for example associated with increasing age or underlying disease (14-16).

In **Chapter 3** we used detailed data of 651 patients to estimate direct medical costs associated with an ICU admission for sepsis. Mean total costs per day were €2,250, cumulating to almost €30,000 for a single ICU admission. Of these costs, the majority was related to accommodation (74%), whilst only 12% was related to diagnostic procedures. Furthermore, using information on patient and disease characteristics available upon presentation to the ICU, we were able to explain only 11% of the variance in health care costs. These findings imply that potential cost- saving strategies based on patient profiling at ICU admission will be limited, and that a diagnostic test such as SeptiCyte LAB will only be cost-effective when significantly reducing ICU length of stay due to earlier initiation of treatment.

Another perspective was chosen in **Chapter 4** in which we combined patient data with data from the Achmea Health Database, which includes all reimbursed health care of more than four million Dutch citizens. Using these data, we were able to assess health care utilization and associated costs after surviving an ICU admission for sepsis, while correcting for trends in health care costs that were already present before the event. Based on data of 357 included patients, we observed an average €2,300 increase

in monthly health care costs when comparing the period before and after sepsis. This increase was mostly due to hospitalization, long-term (home) care, and mental health care. These findings indicate that the economic burden of sepsis reaches far beyond initial ICU treatment. In addition, our study suggested that higher health care expenditure was correlated with diminished health-related quality of life.

Considerations

For both studies examining health care costs, we focused exclusively on patients admitted to the ICU with sepsis. As a result, we could not compare our cost estimations with expenditures related to other admission diagnoses and a conclusion about attributive costs of a nosocomial infection is precluded.

Recently, it has been doubted whether sepsis leads to increased long-term mortality when compared to other acute diseases (17) and studies suggest that long-term outcomes do not differ between groups of ICU survivors but that the long-term consequences of sepsis have been overestimated due to flaws in study design (18, 19). In contrast, studies describing opposite results have also been published (20, 21). These contrasting findings illustrate the methodological challenge of studying long-term sequelae in specific patient populations and the difficulty to select adequate control groups. In **Chapter 4** we tried to avoid these problems by using the patient as his own control, to ascertain that observed trends in resource use were not dependent on pre-existing patient characteristics, such as age, premorbid conditions, or underlying disease processes. Nonetheless, one could not rule out – using an observational study design – the influence of subsequent non-related events during follow-up.

We estimated health care expenditure in patients from two Dutch university hospitals. As there are large differences in health care systems between countries, we cannot generalize our findings to other countries. Nevertheless, our cost estimates related to the acute phase of the disease as described in **Chapter 3** are similar as those reported from other Western European countries (14, 15). Furthermore, although exact estimates might not be similar for other countries due to differences in health care systems and reimbursement policies, the observed change in health care resource use after a sepsis episode as observed in **Chapter 4** is not expected to differ. Thus, we expect that costs will also follow a similar pattern after surviving an ICU admission for sepsis.

Touching upon some ethical aspects is inevitable when describing research on health care costs. From a societal perspective there is a tendency to assign higher monetary value to a treatment if the individual would be worse off without receiving treatment (22). According to the public opinion, the right to protection of life should be prioritized regardless of quality of life, as long as the possible beneficiaries themselves have the desire to live (23). Thus, although costs associated with sepsis are substantial and press available health care budgets, it is unlikely that these expenditures impact societal opinions about ICU treatment for this population.

In health policy making, the quality-adjusted life year (QALY) is used as a measure to compare the effect of different interventions in different settings, by different clinicians, in different subgroups of patients, suffering different conditions, with different disease severity (24). When comparing interventions, the treatment option with the lowest costs and the greatest beneficial effect on QALYs is favoured. In situations such as sepsis requiring ICU admission, it might be valid to compare the situation after an intervention with the situation that an intervention would not have taken place: the so-called without-intervention-utility-level (23). In the Netherlands, a maximum of €80,000 per gained QALY is considered cost-effective. Considering the average health status of 0.70 in ICU survivors of sepsis reported in **Chapter 4**, the maximum acceptable expenditure for ICU treatment of sepsis would be €56,000 in the first year. Keeping in mind the costs of an ICU admission for sepsis and the increase in monthly health care costs after sepsis as estimated in **Chapter 3** and **4**, the threshold per QALY is likely to be exceeded using a time horizon of one year after ICU admission.

In light of the ageing population and worldwide increasing problems such as obesity and comorbid diseases, and the – almost unlimited – medical possibilities, scarcity in health care budgets is evident and costs cannot be ignored when taking decisions in medical processes. Hence, for health policy makers and physicians it will be worthwhile to consider expected health care expenditures, in combination with prognostications of long-term health outcomes and health-related quality of life. The responsibility to initiate this primarily lies by physicians and regulating authorities, as such rational reflections cannot be expected from patients or their families in a period of uncertainty about their future. Transparency about costs associated with treatment towards patients and their families can help in the discussion whether or not to (dis) continue treatment, although prudency is required to prevent that such considerations are solely based on economic elements.

Future perspectives and implications

- The majority of the costs of an ICU admission for sepsis are due to expenditures on personnel, residence, and general disposables per day in the ICU. Diagnostic procedures and therapeutic interventions only comprise a quarter of all costs. Therefore, only strategies that could effectively reduce ICU length of stay could lead to a decrease in costs.
- Studies should be performed to determine society's willingness to pay, physicians' attitudes, and acceptance of patients to make informed decisions regarding discontinuation of ICU treatment based on projected resource use, in combination with expected prognosis on long-term health and health-related quality of life.

PART III. MODELLING LONG-TERM SEQUELAE

As concluded from **Chapter 4,** survivors of sepsis have an increased need for health care, associated with high health care expenditure. Furthermore, a higher dependency on health care was associated with lower health-related quality of life. Sepsis is repeatedly described to be a risk factor for poor outcome after critical illness (20, 25-27). An underlying pathophysiological mechanism could be persisting low-grade inflammation after infection in critical illness, increasing one's susceptibility to develop recurrent infections or (inflammatory-related) chronic diseases (25). In the last part of this thesis, the associations between inflammation during ICU admissions and some of these late consequences – potentially having large impact on society causing economic and disease burden – were explored.

Critical illness-induced hyperglycaemia is frequently observed in patients experiencing severe inflammation and has repeatedly been described as a prognostic factor in the development of type 2 diabetes mellitus (DM2) after ICU stay (28, 29). As we were interested in etiologic factors of the development of DM2, we studied the association between severe inflammation and tissue hypoxia during critical illness and DM2 in the year after ICU discharge in **Chapter 5** using a nested matched case-control study design. During a follow-up period of approximately 14 months, we observed 78 incident cases of DM2 in a cohort of 3185 ICU survivors, which is twice as high as the annual incidence in the age and gender matched general population. Also, our study results suggest a causal relation between hyperinflammation and DM2, but for tissue hypoxia no such association was found.

In **Chapter 6** we studied whether patient and disease characteristics are prognostic factors in developing chronic pain after ICU stay. Newly-acquired chronic pain was reported by 18% of the 1368 patients who survived the first year after ICU discharge. In these patients, the location of pain was highly variable, and the average pain intensity was moderate. We observed neuropathic pain characteristics in half of the patients. Based on our multivariable prediction model, women and patients experiencing severe inflammation during ICU stay had a higher risk of developing chronic pain after ICU discharge. Yet, newly-acquired chronic pain after ICU discharge appeared only moderately predictable.

In critically ill patients with sepsis requiring ICU admission the causes of mortality are many and complex. This is not only true for short-term but also for long-term mortality, up to years after ICU discharge (30). When studying long-term mortality (i.e., time-to-event data), Cox-regression models are frequently used. **Chapter 7** describes a methodological pitfall when using this model, namely the consequences of violation of the proportional hazards assumption (31). Using clinical and simulated data on the association between acute kidney injury and one-year mortality, we demonstrated that non-proportional hazards can be caused by the determinant of interest, confounding

variables, as well as risk factors for the outcome under study. Not taking into account the heterogeneity in the study population introduced by these mechanisms might lead to an over- or underestimation of the true effect.

Considerations

For the studies described in **Chapters 5** and **6** of this thesis we based the primary outcome on a self-reported follow-up questionnaire sent to patients who were still alive one year after ICU discharge. In both **Chapter 5** and **6**, response to the follow-up questionnaire was approximately 70%, thereby allowing the risk of bias by non-response. We did not assess differences in patient characteristics between responders and non-responders, but based on previous studies using this questionnaire – including our research described in **Chapter 4** – it is likely that non-responders were younger, and that there were some differences in premorbid condition (32). In **Chapter 5** we used an exploratory approach, as it was the first study to relate inflammation during critical illness to the subsequent development of diabetes, and we did not formally address non-response bias. This study should thus be considered as hypothesis generating and further research should confirm our findings. In **Chapter 6** we used multiple imputation techniques to replace missing values in the outcome, eliminating this bias by non-response. Although multiple imputation provides unbiased estimates and increases precision and power of study results (33), it may complicate interpretation for intended clinical readers.

Severe inflammation is commonly observed in patients with sepsis, though it is not specific for infection as it is also seen after major surgery, trauma, and burns (34). As described in **Chapters 5** and **6**, for both the development of DM2 and chronic pain after ICU discharge, severe inflammation during ICU admission was an independent risk factor. Although this does not necessarily imply a causal relationship, it may explain why survivors of sepsis have a higher risk of developing late consequences of critical illness than ICU patients with other diagnoses. Indeed, persisting low-grade inflammation following sepsis has been reported previously, and could make subjects more susceptible to late sequelae related to inflammatory processes, such as diabetes and neuropathic pain (35). However, for both studies we operationalized hyperinflammation as the number of days with a CRP>100 mg/L. We did not examine whether this definition was robust and indeed representing a state of severe inflammation as evidenced by other known biomarkers, for example procalcitonin or interleukin-6.

In this thesis, some methodological challenges in sepsis research are briefly discussed, for example diagnostic misclassification and bias by non-response. It goes beyond the scope of this chapter to extensively discuss these challenges and possible solutions, but let the main message be that it is essential to keep in mind methodological pitfalls surrounding statistical methods when performing and reading scientific studies. One of these pitfalls is extensively discussed in **Chapter 7** of this thesis; the risk of

non-proportional hazards in survival data and its impact on the estimated effect using Cox-regression models. This can occur regularly in research involving sepsis patients, as this population is highly heterogeneous (36-38). With our study, we aimed to create awareness amongst clinical researchers and physicians about this subject, so that in both designing and reading a study this challenge might be considered and tried to overcome using statistical solutions.

Future perspectives and implications

- Future research should focus on the evaluation of multidisciplinary strategies for preventing long-term sequelae of critical illness, for example by active follow-up of patients in whom severe inflammation was observed during ICU admission.
- Poor predictability of long-term outcomes after critical illness might limit the possibilities to come up with cost-effective preventive or screening strategies for ICU survivors.

Concluding remarks

Based on this thesis, we conclude that the consequences of sepsis and inflammation are not only evident during an ICU admission, but also beyond ICU discharge and that impact on society is substantial. In the foreseeable future, both the diagnosis and long-term prognosis of sepsis will remain highly uncertain for individual patients, hampering the development of more cost-effective care.

Of course, societal impact comprises more aspects than the ones dealt with in this thesis. Besides the impact of sepsis and inflammation during critical illness on medical costs and late consequences for health, the impact on costs due to sick leave, informal care, life satisfaction, social relations, and ability to work cannot be ignored. For future studies, consultation of the most important stakeholders – survivors of critical illness – should be used to determine which patient-centered outcomes have to be included to cover all aspects of societal impact.

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Appendices

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Nederlandse samenvatting

Infecties zijn één van de meest voorkomende oorzaken van sterfte en gaan gepaard met een hoge ziektelast. Soms reageert het lichaam niet adequaat op een infectie waardoor sepsis kan ontstaan: levensbedreigend orgaanfalen als gevolg van een infectie ergens in het lichaam. Voor patiënten met sepsis is het daarom van levensbelang dat zij tijdige toegang hebben tot zorg om de infectie te bestrijden, dit vereist meestal opname op een intensive care (IC).

Het tijdig diagnosticeren van infectie en sepsis in kritiek ziekte patiënten wordt bemoeilijkt door de verschillende verschijningsvormen. De symptomen van sepsis variëren en zijn afhankelijk van de getroffen organen, de locatie van de infectie en de bestaansduur van symptomen op het moment dat de diagnose wordt gesteld. Dit resulteert meer dan eens in een verkeerde diagnose wat kan leiden tot onjuist gebruik van antibiotica of het verlies van kostbare tijd bij de behandeling.

Sepsis is een belangrijk maatschappelijk probleem. Jaarlijks worden er wereldwijd 30 miljoen mensen in het ziekenhuis behandeld voor sepsis, waarvan er 6 miljoen overlijden. In Nederland is sepsis een veelvoorkomende doodsoorzaak, met 3500 sterfgevallen per jaar. Van de patiënten die sepsis overleven ontwikkelt een groot deel (17%) het zogenaamde post-intensive care syndrome (PICS). Dit zijn fysieke, cognitieve en mentale klachten die voorkomen na een IC-verblijf. Omdat risicofactoren voor sepsis samenhangen met leeftijd en de aanwezigheid van chronische ziekten, is de verwachting dat het aantal gevallen van sepsis in de komende jaren zal toenemen. Tegelijkertijd worden de overlevingskansen van een IC-opname voor sepsis groter. Dit betekent dat er steeds meer mensen in de algehele populatie zullen kampen met de gevolgen van sepsis.

Doel van dit proefschrift

Het doel van dit proefschrift was om te bepalen wat de maatschappelijke impact van sepsis en inflammatie is in patiënten tijdens en na een verblijf op de intensive care (IC). Dit is onderzocht door na te gaan of een nieuwe diagnostische test klinisch voordeel op zou kunnen leveren (Deel I), vervolgens is in Deel II van dit proefschrift geschat wat de economische waarde van zo'n test zou kunnen zijn door enerzijds het berekenen van de zorgkosten van een behandeling voor sepsis op de IC en anderzijds door het berekenen van de zorgkosten geassocieerd met het overleven van een behandeling op de IC. Tot slot is onderzocht welke langetermijnuitkomsten de zorgkosten van IC-overlevers zouden kunnen verklaren (Deel III).

Setting

Sommige studies binnen dit proefschrift werden uitgevoerd binnen het Molecular Diagnosis and Risk Stratification of Sepsis (MARS) project. Dit project is gestart in 2010

in zowel het Academisch Medisch Centrum Amsterdam en het Universitair Medisch Centrum (UMC) Utrecht en had als doel om bij te dragen aan het verbeteren van het diagnostisch en prognostisch proces in IC-patiënten met sepsis. Hiervoor werden dagelijks zowel demografische en klinische data verzameld alsmede bloedplasma dat overbleef na routine labonderzoek. Voor één studie in dit proefschrift werd deze data gekoppeld aan data uit de Achmea Health Database. Deze database bevat informatie over verzekerde gezondheidszorg van meer dan 4 miljoen Nederlandse burgers voor de periode waarin ze bij de betreffende verzekeraar waren aangesloten. Naast deze data werd ook informatie gebruikt die werd verkregen middels een vragenlijst die in het UMC Utrecht – voornamelijk voor kwaliteitsdoeleinden – aan alle IC-overlevers wordt verstuurd één jaar na IC-verblijf. Deze vragenlijst bevat onder andere vragen over huidig medicijngebruik, fysiek functioneren, gezondheidsgerelateerde kwaliteit van leven, angst, depressie en pijn.

Deel I. Het diagnosticeren van infectie

Sepsis is geassocieerd met hoge sterfte en ziektelast. Vroegtijdige interventies zouden bij kunnen dragen aan het verlichten van de ziektelast maar dit wordt belemmerd door de moeizame vroege herkenning en diagnose van sepsis. De laatste jaren is van verschillende biomarkers onderzocht of zij bij kunnen dragen aan het diagnosticeren van infectie. Echter, de resultaten van deze studies impliceren dat het gebruik van één enkele biomarker niet voldoende accuratesse oplevert. Daarom wordt momenteel veelvuldig onderzoek gedaan naar meer complexe combinaties van biomarkers. Een voorbeeld van zo'n complexe biomarkertest is de SeptiCyte LAB™ test (Immunexpress, Seattle, WA). Eerdere onderzoeken die verschenen over de diagnostische prestaties van deze test waren veelbelovend. Deze studies waren veelal technisch van aard en maakten gebruik van een sterk geselecteerde populatie.

In **Hoofdstuk 2** wordt een klinische validatiestudie beschreven waarbij de diagnostische en prognostische prestaties van de SeptiCyte LAB test is onderzocht in een cohort van 467 patiënten die vanuit het ziekenhuis opgenomen werden op de IC met acuut respiratoir falen. De referentiestandaard was de prospectieve beoordeling van aanwezigheid van infectie tijdens IC-opname. Hoewel de sensitiviteit van de test hoog bleek (98%), was er een groot aantal foutpositieven wat resulteerde in een specificiteit van slechts 18%. Daarmee was het onderscheidend vermogen van de test matig en niet beter dan één enkele biomarker, die goed beschikbaar is in de dagelijkse praktijk en – naar alle waarschijnlijkheid – een stuk minder kostbaar dan de SeptiCyte LAB test. Vanuit prognostisch oogpunt voegden de resultaten van de test niets toe bovenop het gebruik van een reeds geaccepteerde index die gebruikt wordt bij het voorspellen van ziekenhuismortaliteit (APACHE IV score).

Deel II. Het schatten van economische gevolgen

Zowel de periode van IC-verblijf als de periode na IC-ontslag kan gepaard gaan met hoge zorgkosten. Eerdere studies die de zorgkosten geassocieerd met sepsis hebben onderzocht werden beperkt door het gebruik van administratieve data om sepsis vast te stellen en zorgverbruik te schatten. Dit belemmert het schatten van zorgkosten op het niveau van de individuele patiënt. Andere studies gebruikten slechts een beperkte tijdsperiode om de kosten te schatten, waardoor niet gecorrigeerd kon worden voor trends in zorgkosten, bijvoorbeeld trends geassocieerd met stijgende leeftijd of een onderliggende ziekte.

In **Hoofdstuk 3** gebruikten we gedetailleerde data van 651 patiënten om de medische kosten van een IC-verblijf voor sepsis te schatten. De gemiddelde totale kosten waren €2.250 per dag en bijna €30.000 voor een enkele IC-opname. Van deze kosten was de meerderheid gerelateerd aan accommodatie (74%), terwijl maar 12% gerelateerd was aan diagnostische procedures. De zorgkosten bleken ook moeilijk te voorspellen aan de hand van patiëntgegevens die beschikbaar waren bij IC-opname. Deze bevinding impliceert dat kostenbesparende strategieën gebaseerd op patiëntprofielen beperkt zijn en dat een diagnostische test zoals SeptiCyte LAB alleen kosteneffectief kan zijn als de ligduur van patiënten door vroege en accurate diagnose en behandeling wordt verkort.

Een ander perspectief werd gekozen in **Hoofdstuk 4** waarin patiëntgegevens werden gecombineerd met data van de Achmea Health Database. Met behulp van deze informatie kon het zorggebruik en de bijbehorende zorgkosten na het overleven van een IC-opname voor sepsis in kaart worden gebracht, waarbij werd gecorrigeerd voor onderliggende trends in ziektekosten. Gebaseerd op de gegevens van 357 patiënten werd geconcludeerd dat de gemiddelde zorgkosten met €2.300 per maand stegen in de periode na sepsis wanneer deze werd vergeleken met de periode ervoor. Deze toename werd met name veroorzaakt door opname in een ziekenhuis of verpleeghuis, thuiszorg en geestelijke gezondheidszorg. Deze bevindingen laten zien dat de economische gevolgen van sepsis ver voorbij de initiële IC-behandeling reiken. Ook hingen hoge zorgkosten samen met verminderde gezondheidsgerelateerde kwaliteit van leven één jaar na IC-ontslag.

Deel III. Het modelleren van langetermijnuitkomsten

Overlevers van sepsis hebben dus een verhoogd zorggebruik en hoge zorgkosten; indicaties voor een verminderde gezondheid. In de literatuur is sepsis vaak beschreven als een risicofactor voor een slechte uitkomst na IC-verblijf. Een onderliggend pathofysiologisch mechanisme zou persisterende laaggradige inflammatie na infectie kunnen zijn, waardoor de vatbaarheid voor infecties en chronische ziektes toeneemt. In het laatste deel van dit proefschrift werd daarom de associatie tussen inflammatie tijdens IC-verblijf en een aantal langetermijnuitkomsten – die bijdragen aan hoge zorgkosten – bestudeerd.

Hyperglykemie komt vaak voor tijdens een IC-opname in patiënten met ernstige inflammatie en is herhaaldelijk beschreven als een prognostische factor in het ontwikkelen van diabetes mellitus type 2 (DM2) na IC-verblijf. Voor het onderzoek beschreven in **Hoofdstuk 5** waren we geïnteresseerd in etiologische factoren, daarom is de associatie tussen ernstige inflammatie en weefselhypoxie tijdens IC-verblijf en het ontwikkelen van DM2 in het jaar na IC-ontslag bestudeerd. Hiervoor werd een zogenaamd patiënt-controleonderzoek gebruikt. In de eerste 14 maanden na IC-ontslag ontwikkelden 78 van de 3.185 patiënten diabetes, een incidentie tweemaal zo hoog als de jaarlijkse incidentie in de algehele populatie (vergelijkbaar op leeftijd en geslacht). De bevindingen lijken ook te wijzen op een causale relatie tussen hyperinflammatie en DM2, deze relatie werd niet gevonden tussen weefselhypoxie en DM2.

In **Hoofdstuk 6** werd onderzocht of patiëntkarakteristieken en karakteristieken van het IC-verblijf prognostische factoren zijn van het ontwikkelen van chronische pijn na IC-verblijf. Nieuw verworven chronische pijn werd geobserveerd in 18% van de 1.368 patiënten die het eerste jaar na IC-verblijf overleefden. De locatie van de pijn varieerde sterk en de gemiddelde pijnintensiteit was matig. Karakteristieken van neuropathische pijn werden geobserveerd in de helft van de patiënten. Uit een multivariabel predictiemodel kwam naar voren dat vrouwen en patiënten die ernstige inflammatie doormaakten tijdens IC-verblijf een hogere kans hebben om chronische pijn te ontwikkelen. Echter, als geheel functioneerde het predictiemodel slechts matig.

In kritiek zieke patiënten met sepsis zijn vele oorzaken voor mortaliteit te onderscheiden. Dit is niet alleen het geval als het gaat om kortetermijnsterfte maar dit geldt tot jaren na IC-ontslag. Voor het bestuderen van deze langetermijnsterfte wordt vaak een Cox-regressiemodel gebruikt. **Hoofdstuk 7** beschrijft een methodologische valkuil bij het gebruiken van een dergelijk model, namelijk de consequenties van het niet voldoen aan de assumptie van proportionele hazards. Door middel van klinische en gesimuleerde data van de associatie tussen acuut nierfalen en mortaliteit is het effect van niet-proportionele hazards, veroorzaakt door verschillende mechanismen, beschreven. Hieruit blijkt dat het niet in ogenschouw nemen van (niet geobserveerde) heterogeniteit in de studiepopulatie kan leiden tot zowel een onder- als overschatting van het daadwerkelijke effect.

Slotopmerkingen

Op basis van dit proefschrift kan geconcludeerd worden dat de gevolgen van sepsis en inflammatie niet alleen zichtbaar zijn tijdens een IC-opname maar ook lang na IC-ontslag. De maatschappelijke impact – uitgedrukt in zowel economische gevolgen als ziektelast – is substantieel. In de nabije toekomst zullen zowel de diagnose als langetermijnprognose van sepsis in hoge mate onzeker blijven voor individuele patiënten wat de ontwikkeling van meer kosteneffectieve zorg belemmert.

Natuurlijk omvat de maatschappelijke impact van sepsis en inflammatie meer aspecten dan die zijn opgenomen in dit proefschrift. Naast de impact op zorgkosten en langetermijnuitkomsten is er de impact op kosten door ziekteverzuim en mantelzorg, levensgeluk, sociale relaties en de mogelijkheid om te werken. Voor toekomstige studies is het daarom van uiterst belang om de belanghebbenden – overlevers van sepsis – te consulteren om samen te bepalen welke patiëntgerichte uitkomsten geïncludeerd zouden moeten worden om alle aspecten van maatschappelijke impact te kunnen onderzoeken.

Dankwoord

"That's one small step for mankind, a giant leap for one woman." (Naar Neil Armstrong, 1969)

Zo voelt het voor mij nu ik eindelijk het belangrijkste deel – het dankwoord – van dit proefschrift kan schrijven. Het is klaar! Het is geen valse bescheidenheid als ik zeg dat ik dit zonder (mentale dan wel praktische) steun van heel veel mensen niet had kunnen doen. Daarvan wil ik er een aantal bedanken. Een disclaimer daarbij is dat het niet mogelijk is om iedereen bij naam te noemen. Maar als je tot dit rijtje behoort dan wéét je dat en ben ik je daar eeuwig dankbaar voor.

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Curriculum Vitae

Marlies Brouwer werd 23 mei 1989 geboren te Urk, als zesde kind van wijlen Jan Brouwer en Riek Brouwer-Bonnink. In 2006 behaalde zij haar havodiploma aan de Pieter Zandt scholengemeenschap en in 2010 rondde zij haar studie verpleegkunde af aan de Christelijke Hogeschool Ede. Na haar diplomering werkte Marlies als wijkverpleegkundige, eerst bij Zorggroep Oude en Nieuwe Land (2010-2011), later bij Icare (2011-2012).

Gedurende haar hbo-opleiding raakte Marlies geïnteresseerd in het doen van onderzoek. Daarom volgde zij naast haar baan de premaster Health Sciences aan de Vrije Universiteit Amsterdam. Na het afronden van de premaster startte zij met de research master Lifestyle and Chronic Disorders aan dezelfde universiteit. Hiervan behaalde zij in 2014 haar diploma.

In het tweede jaar van haar research master deed Marlies afstudeeronderzoek binnen het Molecular Diagnosis and Risk Stratification of Sepsis (MARS) project in het UMC Utrecht. Dit onderzoek mondde uit in een promotietraject onder supervisie van dr. Olaf Cremer en prof. dr. Marc Bonten bij het Julius Centrum voor Gezondheidswetenschappen en Eerstelijns Geneeskunde. Tijdens haar promotieonderzoek behaalde Marlies de postgraduate master Epidemiology met als specialisatieprogramma Medical Statistics aan de Universiteit van Utrecht.

In augustus 2018 startte Marlies als docent-onderzoeker Gezondheid & Technologie bij hogeschool Saxion te Deventer. De komende jaren hoopt zij zich verder te ontwikkelen binnen het hoger onderwijs.

Marlies is in 2012 getrouwd met Jan Willem Koster. Sindsdien wonen zij in Kampen. Samen hebben zij een zoon, Gideon (2017).