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INVESTIGATION OF TOLL-LIKE RECEPTOR 4 LEVELS IN PATIENTS WITH RESPIRATORY INFECTIONS WITH INDIVIDUALIZED INFUSION AND ANTIMICROBIAL MANAGEMENT

Výskum úrovní Toll-like receptorov 4 u pacientov s respiračnými infekciami s personifikovaným tekutinovým a antimikrobiálnym manažmentom

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Abstract
The aim of the study is to evaluate the dynamics of TLR-4 levels and microbial colonization in patients with infectious respiratory complications with different treatment strategies.
Materials and Methods. 20 children in each group with different approaches to treatment - in the main observation group in the complex of antimicrobial treatment at the same time with empirically prescribed standard antibiotic therapy we used inhalation of quaternary ammonium compound (decamethoxine) through a nebulizer from the appearance of clinical signs of VAP, and the treatment of patients in the control group was carried out in accordance with generally accepted recommendations with the use of systemic antibacterial and symptomatic therapy.
Results. It was found that the level of TLR-4 in the serum clearly correlates with the dynamics of the infectious process and the microbiological status of patients.
Conclusions. The determination of the level of TLR-4 can serve as a diagnostic criterion for the development of severe bacterial disease and evaluate the effectiveness of the selected treatment regimen (Fig. 6, Ref. 60). *Text in PDF www.lekarsky.herba.sk.*
KEY WORDS: VAP, children, toll-like receptors 4, quaternary ammonium compound, decamethoxine.

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Introduction

Ventilator-associated respiratory infection (VARI) is a common hospital complication in the intensive care unit associated with medical care (49, 58, 59). The disease includes ventilator-associated tracheobronchitis (VAT) and ventilator-associated pneumonia (VAP). The incidence in developed countries is 1-22 cases per 1000 days of ventilation (18, 20, 37). VAP occurs among 20 % of patients who are on mechanical ventilation for more than 48 hours. The mortality rate reaches 30 % (8, 12, 13).

It has been proven that VARI are associated with increased treatment costs and length of stay in intensive care (48, 53, 54). It is difficult to estimate the mortality rate associated with VARI because patients who require prolonged ventilation are often seriously ill and have comorbid conditions. Studies indicate that respiratory infections associated with medical care are mainly caused by gram-negative organisms resistant to many antimicrobial drugs (26, 42, 52). Some studies indicate unfavorable survival rates of this category of patients (4, 5, 38). The association between high morbidity and antibiotic resistance suggests that the choice of antimicrobial treatment for VARI is important. From these positions, the identification of factors associated with the

early determination of the etiological factor will increase the effectiveness of prevention and treatment (4, 17, 57). Therefore it is of great clinical importance to monitor and detect even slight changes occurring in the spectrum of microorganisms that cause complications, depending on the types of surgical intervention, its location, and some others (15, 55, 56).

VAP refers to nosocomial infections associated with the provision of medical care with the highest mortality rate among surgical patients (11, 47). Methods and criteria for the diagnosis of VAP are still controversial (7, 33).

It is known that only approximately 1/3 - 1/4 of patients with suspected VAP have been verified microbiologically (10, 21, 39). Clinically, the diagnosis of VAP is often determined in the intensive care unit, accompanied by the appointment of empirical, not always justified, antibiotics (32, 41, 46). In recent years, scientists have developed new approaches to identifying biological markers that can accurately and quickly diagnose VAP. Among these are known toll-like receptors (TLR) (19, 23, 53).

Toll-like receptors recognize pathogens and generate an immediate protective response by inducing the production of pro-inflammatory cytokines (45). TLRs are expressed in different cell types of the immune system and are associated with different states of cellular activation, immune protection, maintenance of homeostasis and various diseases (35). Toll-like receptors of type 4 (TLR-4) specifically recognize bacterial lipopolysaccharide, and its activation leads to the synthesis of pro-inflammatory cytokines and chemokines (29, 30). TLR-4 and related immunological pathways are now widely studied for diagnostic and therapeutic purposes in infectious complications, including respiratory system.

The relationship between fluid balance (FB) and ventilator-associated events (VAEs) has only been investigated in a few studies with small sample sizes and is not fully understood (9, 34, 36). As an important approach to optimize tissue perfusion, fluid therapy is widely used to improve organ perfusion and survival in patients with critical illness (1, 44, 57). Inadequate fluid resuscitation is associated with poor prognosis; the former may lead to tissue hypoperfusion and exacerbate organ dysfunction, whereas the latter can increase the risk of heart failure, pulmonary edema, and pleural effusions (6, 9, 31). Optimal infusion therapy is difficult to achieve with standard parameters (e.g., heart rate (HR), blood pressure (BP), central venous pressure (CVP)) (34, 40, 56). In recent years, various non-invasive hemodynamic monitoring technologies have been proposed (14, 50). Innovative technologies for continuous non-invasive hemodynamic monitoring significantly expand the possibilities of improving the strategy of infusion therapy and personalization of hemodynamic management (14, 51).

The aim of the study was to evaluate the dynamics of TLR-4 levels and microbial colonization in patients with infectious respiratory complications with different treatment strategies and fluid management based on non-invasive hemodynamic monitoring indicators.

Material and methods

The studies were conducted with the obligatory information of the parents of the patients and with their written consent in accordance with the measures concerning the safety of the patient's health, respect for human rights, human dignity and moral and ethical norms provided by the principles of the Helsinki Declaration of Human Rights, human and biomedicine, relevant laws, orders of the Ministry of Health of Ukraine. Our study was approved by the local Bioethics Committee (protocol 5, No. 0122U002551, 2022).

The study involved 40 children who were on mechanical ventilation (MV) for 48 hours or more. All examined patients were 8-16 years old. There were 22 (55 %) girls and 18 (45 %) boys. Infectious airway complications were diagnosed as ventilator-associated pneumonia according to all criteria according to the US Centres for Disease Control and Prevention (CDC). According to these recommendations, the diagnosis of VAP was based on clinical symptoms, radiographic criteria with mandatory microbiological confirmation (22, 25).

The main exclusion criteria for participation in this study were the following: patients with acute kidney injury or chronic kidney disease accompanied by anuria or oliguria, patients who has comorbid pathologies (e.g., heart failure, diabetes mellitus or other acute conditions), one of the parents did not provide a consent for the child's participation in the clinical trial.

All patients received standard comprehensive treatment according to the treatment protocols of the underlying pathology. Respiratory support was performed with a ventilator "Hamillton-C2" in ASV mode.

Empirical antibacterial therapy was consistent with conventional approaches to the treatment of VAP and included drugs in accordance with the treatment protocol for this pathology.

The hemodynamic effects of the treatment strategies and the need for infusion therapy were assessed by continuous non-invasive measurement of estimated continuous cardiac output (esCCO) and cardiac index (esCCI) by a Nihon Kohden monitor and were calculated based on pulse oximetry data, ECG signals, and pulse wave transit time (27).

Patients were divided into two groups of 20 people each with different approaches to the treatment of VAP. In the main observation group, inhalation of decamethoxine (DCM) through a nebulizer from the first day after the appearance of clinical signs of VAP was used simultaneously with the empirically prescribed standard antibiotic therapy. DCM is a surfactant bisquaternary ammonium compound consisting of a synthetic decamethylene molecular moiety and menthol (L-menthol) peppermint oil (1,10-Decamethylene bis (N, N-dimethylmethoxycarbonylmethyl) ammonium dichloride). Inhalations of 5 ml of DCM solution were per-

formed daily for 7 days three times a day through a nebulizer, which was connected to the patient's respiratory circuit at the inspiratory end 15 – 20 cm above the special „Y-shaped" convector. Treatment of patients in the control group was performed in accordance with generally accepted recommendations with the use of systemic antibacterial and symptomatic therapy.

The effectiveness of treatment was assessed by clinical indicators of the disease, data from microbiological examination of tracheobronchial secretion and the level of TLR-4 in the serum. Clinically, the effectiveness of treatment of patients in the DCM and control groups was determined by general clinical indicators of the patient's condition (body temperature, self-breathing efficiency, blood oxygen saturation), laboratory (peripheral blood leukocyte content) and instrumental (lung X-ray) indicators.

The microbiology analysis

The microbiological study consisted of isolating clinically significant pathogens in a pure culture in patients with infectious respiratory complications (VAP) and then identifying them by morphological, tinctorial, cultural, and biochemical properties to determine their taxonomic position. Biochemical properties of isolated strains of gram-positive and gram-negative microorganisms, their species identification were determined using standardized test systems STAFI test-16, ENTERO test-24, and NEFERM test-24 (PLIVA – Lachema a. S. Brno, Czech Republic). Final identification was performed using an automatic bacteriological analyzer Vitec - 2compact bioMérieux (France).

Simultaneously with the determination of the species of pathogens of infectious complications, the quantitative determination of microorganisms in the biological material obtained from patients with VAP was performed according to standard methods. The degree of total microbial colonization of the respiratory tract was assessed by the number of microorganisms in 1 ml of material, expressed in log CFU/ml in the laboratory on the basis of a research bacteriological laboratory of the Department of Microbiology, Vinnytsya National Pirogov Memorial Medical University, certified by the Ministry of Health of Ukraine.

TLR-4 determination

Assessment of serum TLR-4 was performed in patients on the 1st, 3rd, 7th day of treatment. Determination of TLR-4 was performed by TLR-4 in serum enzyme-linked immunosorbent assay, according to the instructions of the manufacturer of the set "Human TLR4 ELISA Kit" (NeoBiolab, USA). The essence of the method was that in the wells of the tablets, on the walls of which are adsorbed antibodies to estradiol, added 100 µl of standard solutions (with estradiol concentrations – 0; 1,0; 2,5; 5,0; 10,0; 25, 0 ng/ml), 50 µl of enzyme conjugate (streptavidin peroxidase), stirred for 10 s. We incubated for 60 min at 37 °C in a humid chamber to form a solid phase complex AT-AG-AT enzyme. The wells were washed of excess unbound reagents and 50 µl of the chromogenic substrate was added. Stirred, incubated for 15 minutes at 25 °C, the reaction was stopped with 50 µl of stop solution and photometered at 450 nm (differential filter 630 nm) on an automatic analyser STAT FAX 303/PLUS. Serum samples stored at –20 °C in Eppendorf microtubes. All samples are suitable for research. Hemolyzed, lipemic serum samples and coagulated samples were not examined. Research of biological material was carried out at the Department of Biological and General Chemistry in the research clinical and diagnostic laboratory of National Pirogov Memorial Medical University, Vinnytsya (certificate of the Ministry of Health of Ukraine on re-certification No. 049 /15 dated 02.03.2015).

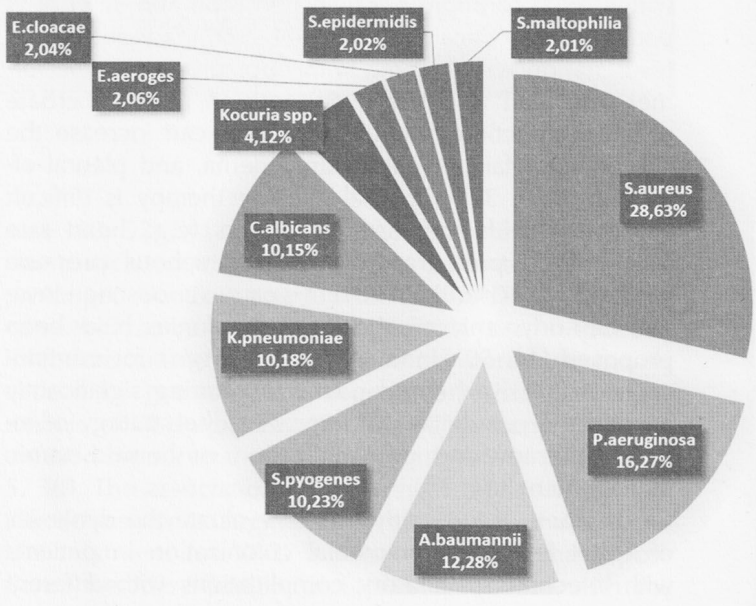
Statistical analysis

The results were statistically processed using Microsoft Excel 2016 and "Statistica 5.5" (license No AXXR910A374605FA). Probability analysis was performed according to Student's t-test. The r-Pearson test was determined to assess the presence and strength of the association between the symptoms. Results at $P<0.05$ were considered statistically significant. In addition, multivariate analysis of variance (MANOVA) with a probability less than 5 % was used to determine the effect of the selected treatment regimen on microbial colonization of the respiratory tract, serum level of TLR-4, esCCO and daily infusion volume.

Results

As a result of the microbiological examination, 40 samples of biological material obtained from patients (n = 40) with VAP in the first days after the onset of the disease were isolated and identified by morphological, tinctorial, cultural, and biochemical properties of 52 strains of opportunistic pathogens (Fig. 1).

Figure 1. Etiological structure of pathogens from the total number of strains of microorganisms (n = 52) **isolated from critically ill patients with VAP at the beginning of treatment.**



In patients of both groups among 52 strains of isolated microorganisms, the leading place was occupied by both Gram-positive and Gram-negative pathogens: *S. aureus* (28.63 %), *P. aeruginosa* (16.27 %), *A.baumannii* (12.28 %), *S. pyogenes* (10.23 %), *K. pneumoniae* (10.18%), *C. albicans* (10.15 %).

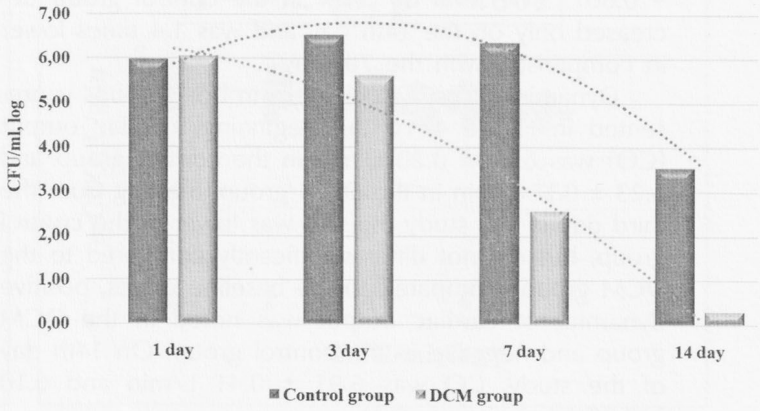
Microbiological examination of biological material obtained from the airways of patients showed that at the beginning of treatment the average values of the number of microorganisms, expressed in log, in the DCM group (6.65 ± 0.29 CFU/ml) did not differ significantly from the degree of microbial colonization of the airways patients of the control group (5.9 ± 0.22 CFU/ml; $p > 0.05$). During treatment, a gradual decrease in the quantitative content of microorganisms in the test material obtained from patients of both groups was determined microbiologically. In DCM group a significant decrease in microbial colonization was found starting from the third day of treatment (log (5.65 ± 0.24 CFU/ml) (Fig. 2).

Microbial colonization of the airways in patients of the control group on the third day was 1.1 log higher, which indicated a predominance of 14 times the absolute number of microorganisms in the airways of patients in the control group ($p < 0.001$). When using decasan in patients there was a significant decrease in the number of microorganisms in the secretions from the tracheobronchial tree by 3 orders of magnitude (log (2.72 ± 0.38) CFU/ml) compared with the initial level of microbial colonization ($p<0,001$), which in absolute terms corresponded to a decrease in CFU of 1 ml by 104 times. Such values of microorganisms in the test material did not exceed the critical level of microbial load, which is in decimal logarithms 4 CFU/ml. In the sputum of most patients the DCM group of pathogens of infectious complications after 14 days was not isolated.

Patients in the control group who received only standard systemic antibiotic therapy had higher rates of airway colonization during the first week of treatment. On the seventh day, the number of opportunistic pathogens identified in these patients with VAP reached log (6.49 ± 0.28) CFU/ml, which is four orders of magnitude higher than the content of microorganisms in the inflammatory focus in patients of DCM group ($p < 0.001$).

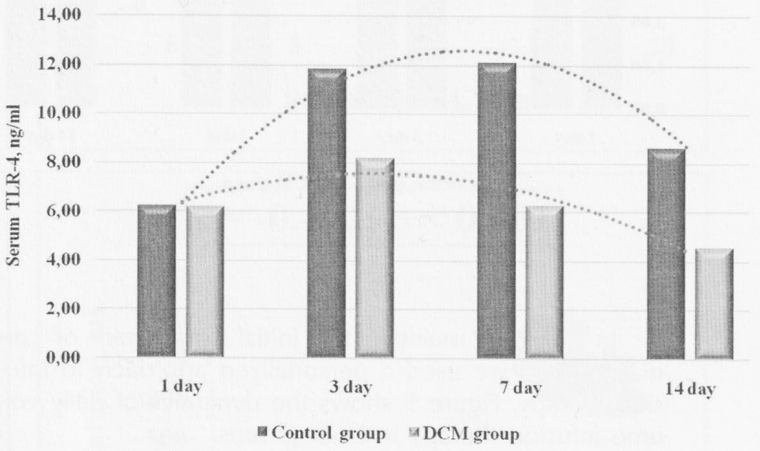
A significant decrease in the number of microorganisms in the tracheobronchial secretion of patients in the control group was observed not earlier than after 10 days of receiving systemic antibiotic therapy. Thus, after 14 days it was found that the total number of microorganisms in 1 ml of tracheobronchial aspirate expressed in log decreased almost twice compared to the seventh day, and was (3.54 ± 0.21) CFU/ml, which in 18 times significantly exceeded this figure in DCM group ($p < 0.001$) (Fig. 2).

Figure 2. Indicators of microbial colonization of the respiratory tract of patients with VAP in both groups. The average values of the total number of microorganisms in 1 ml of tracheobronchial aspirate are expressed in log (CFU/ml).



When used in patients with VAP inhalation of antiseptic drug based on DCM was found less increase in serum TLR-4 in the early period from the start of treatment. Thus, on the 3rd day in patients of the DCM group noted 1.45 times lower level of TLR-4 than in the control group ($p < 0.001$) (Fig. 3).

Figure 3. Serum levels of TLR-4 in patients with VAP (in ng/ml).



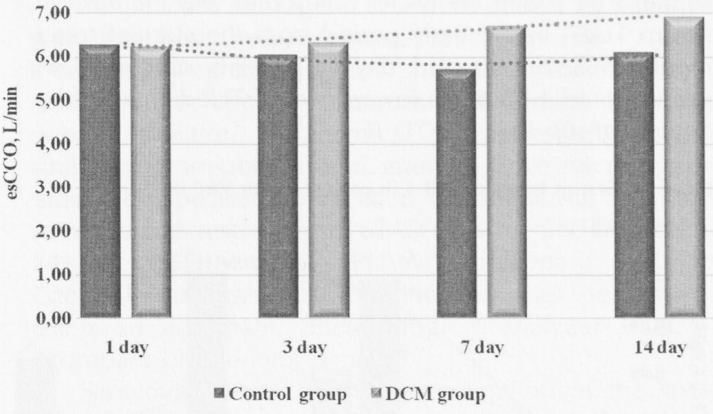
In the control group, a significant increase in 1.9 times in serum levels of TLR-4 ($p < 0.001$) was found at the same time as the increase in colonization by gram-negative bacteria in the tracheobronchial aspirate on the 3rd day (r-Pearson = 0.880).

The positive dynamics of the decrease in the number of microorganisms in the aspirate obtained from the patients of DCM group was marked by a decrease in the level of TLR-4 on the 7th and 14th day of treatment. Evidence of high efficacy of additional use of DCM in the complex treatment of patients with VAP is a strong correlation between a decrease in gram-negative pathogens of respiratory infections and a decrease in TLR-4 to almost baseline (r-Pearson = 0.893). In comparison, the level of TLR-4 in the serum of patients with VAP who received only systemic therapy remained twice as

high (p <0.001) than in the DCM group, and correlated with inflated levels of gram-negative microorganisms in the tracheobronchial aspirate of patients (r-Pearson = 0.801). The level of TLR-4 in the control group decreased only on the 14th day and was 1.4 times lower in comparison with the 7th day.

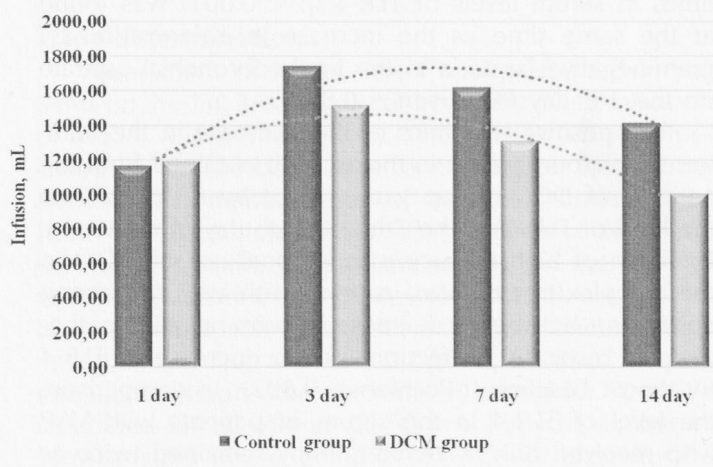
Dynamics of esCCO changes in both groups is presented in Figure 4. At the beginning, cardiac output (CO) was 6.27 ± 0.20 L/min in the control group and 6.23 ± 0.17 L/min in the DCM group. Starting from the third day of the study the CO was lower in the control group, but did not differ significantly compared to the DCM group. Compared to the baseline values, positive dynamics of cardiac output was noted in the DCM group and negative in the control group. On 14th day of the study, CO was 6.91 ± 0.41 L/min and 6.10 ± 0.39 L/min, respectively.

Figure 4. Cardiac output dynamics by estimated continuous cardiac output (esCCO) **using non-invasive monitoring.**



In order to maintain the initial parameters of cardiac output, we used a personalized approach to infusion therapy. Figure 5 shows the dynamics of daily volume infusion therapy in both groups.

Figure 5. The dynamics of daily volume infusion therapy in both groups.



At the beginning of treatment, the infusion volume was almost the same in both study groups: 1165 ± 108,9 ml/day in the control group and 1190 ± 137.3 ml/day in the DCM group. From the 3rd day the infusion volume increased in both study groups, but patients in the control group required significantly higher infusion volume compared to the DCM group. On 14th of the study, the infusion volume in the control group was higher than at the beginning of treatment and amounted to 1415 ± 130.9 ml/day, opposite to the DCM group where the infusion volume decreased to 1005 ± 82.6 ml/day.

The results of the multivariate analysis of variance (MANOVA) on the 3rd day of the study show that the treatment regimen with the additional inhalations of Decametoxin has a reliable effect on the set of the leading investigated indicators (CFU, Serum TLR-4, esCCO, daily infusion volume – $F(4.35) = 16.658$; $p < 0.0001$; Wilk's $\Lambda = 0.344$). The results of the MANOVA also demonstrated the advantages of the applied technique on the 7th day of the study ($F(4.35) = 67.162$; $p < 0.0001$; Wilk's $\Lambda = 0.115$), and on the 14th day of the study ($F(4.35) = 117,312$; $p < 0.0001$; Wilk's $\Lambda = 0.069$). It should be noted that the reliability of differences between groups increased over time.

Correlations of all studied parameters are presented in Figure 6.

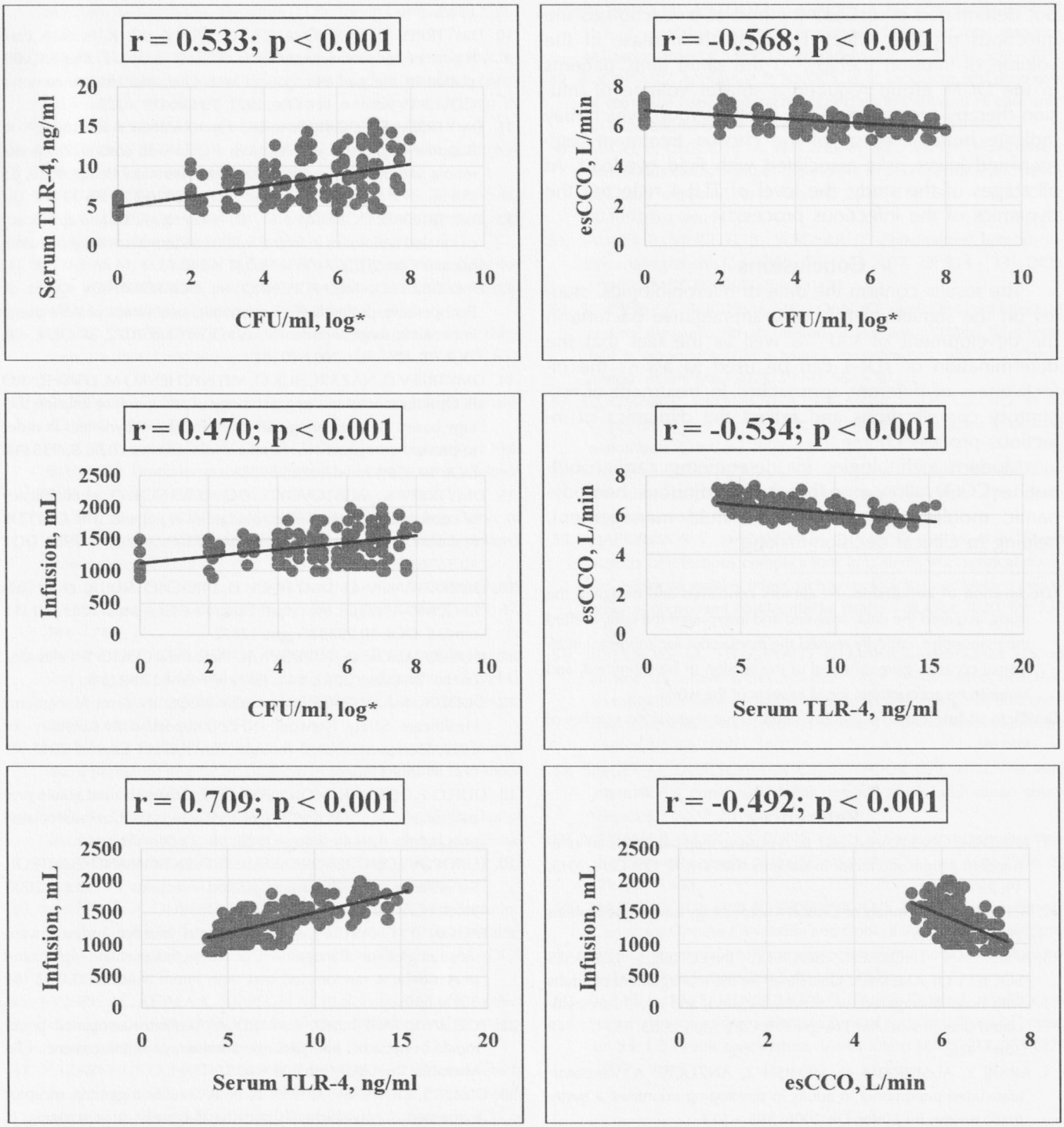
Discussion

It is known that VAP occurs during prolonged invasive mechanical ventilation, usually within 48 – 72 hours. There are signs of systemic infection - fever, changes in white blood cell count, changes in the nature of sputum, and deterioration of the radiological picture of the lungs (3, 16). The frequency of diagnosis of VAP depends on the microbiological status of the patient, the microbiological passport of the department, the length of stay of the patient on mechanical ventilation, antibiotic regimens, and approved local criteria for diagnosis (2). Differences in diagnostic criteria, as well as ambiguous approaches to the appointment of antibiotic therapy, lead to an increase in the emergence of multidrug-resistant pathogens (28, 43, 60).

Currently, the development and improvement of new antimicrobial drugs for the treatment of VAP is significantly progressing, but clear diagnostic criteria have not yet been developed, which often leads to irrational antibiotic therapy and the development of multidrug-resistant pathogens. The level of TLR-4 we determined as an additional diagnostic criterion in the diagnosis of VAP. Determination of TLR-4 levels on days 1, 3, 7 and 14 from the beginning of treatment served as a marker of rational antibiotic therapy, reflected the dynamics of the infectious process and allowed to evaluate the effectiveness of the selected antimicrobial treatment regimen.

Among critically ill patients, adequate fluid resuscitation is essential for restoring CO and renal perfusion, however, excess fluid can lead to cardiopulmonary complications including pulmonary edema and conges-

Figure 6. Correlations of all studied parameters. 1 – moderate positive relationship; 2 – moderate negative relationship; 3 – moderate positive relationship; 4 – moderate negative relationship; 5 – strong positive relationship; 6 –moderate negative relationship.



tive heart failure (31), resulting in impaired gas exchange and contractility, conduction disturbance, reduced compliance, and diastolic dysfunction. Indeed, 20 – 40 % of VAEs may be attributable to congestive heart failure and pulmonary edema caused by fluid overload (31, 34). In addition, patients with pulmonary edema and impaired respiratory function are also more susceptible to bacterial infection (12, 15). The optimal fluid management of intensive care unit (ICU) patients with VAP has not been established. Clinical characteristics vary among ICU patients, making is difficulties to detect fluid overload for individual patients. Therefore, optimizing tissue perfusion in patients with VAP relies on an individualized approach. Goal-directed fluid therapy aims to optimize DO2 by maintaining or increasing cardiac output. This preserves the microbicidal function of immune cells and the protection of organs that are particularly sensitive to perioperative hypoperfusion (24), avoiding intestinal barrier failure and GALT disorders.

The results of our study showed that in patients of both study groups there was a decrease in cardiac output determined by esCCO method as a reaction to the infectious process, which required an increase in the volume of infusion therapy. At the same time, patients in the DCM group required a smaller volume of infusion therapy compared to the control group, which may indicate better efficacy of the chosen treatment regimen and lower risks associated with fluid overload. At all stages of the study, the level of TLR-4 reflected the dynamics of the infectious process.

Conclusions

The results confirm the data of microbiological studies on the significant role of gram-negative bacteria in the development of VAP, as well as the fact that the determination of TLR-4 can be used to assess the effectiveness of different approaches to treatment of respiratory complications and reflect the dynamics of infectious process course.

Modern technologies for determining cardiac output (esCCO) allow non-invasive continuous hemodynamic monitoring and optimize fluid management, helping in clinical decision-making.*

*__Declaration of authorship.__ All authors conceived and designed the study, acquired the data, analyzed and interpreted the data, drafted the manuscript, critically revised the manuscript for important intellectual content, gave approval of the version to be submitted, and agree to be accountable for all aspects of the work.

__Conflicts of Interest.__ The authors declare that there is no conflict of interest.

References

1. ACHEAMPONG A, VINCENT JL. A positive fluid balance is an independent prognostic factor in patients with sepsis. Crit Care 2015, 19: 251.
2. ALP E, VOSS A. Annals of Clinical Microbiology and Antimicrobials 2006, 5: 7.
3. AMERICAN THORACIC SOCIETY, INFECTIOUS DISEASES SOCIETY OF AMERICA: Guidelines for the management of adults with hospital-acquired, ventilatorassociated, and healthcare-associated pneumonia. Am J Respir Crit Care Med 2005, 15, 171 (4): 388 – 416.
4. ARABI Y, AL-SHIRAWI N, MEMISH Z, ANZUETO A. Ventilator-associated pneumonia in adults in developing countries: a systematic review. Int J Infec Dis 2008: 505 – 512.
5. BLOT S, KOULENTI D, DIMOPOULOS G, et al. Prevalence, risk factors, and mortality for ventilator-associated pneumonia in middle-aged, old, and very old critically ill patients. Crit Care Med 2014, 42 (3): 601 – 609.
6. BOYD JH, FORBES J, NAKADA TA, et al. Fluid resuscitation in septic shock: A positive fluid balance and elevated central venous pressure are associated with increased mortality. Crit Care Med 2011, 39: 259 – 265.
7. CHASTRE J, FAGON JY. Ventilator-associated pneumonia. Am J Respir Crit Care 2002, 65: 867 – 903.
8. CHASTRE J, FAGON JY. VAP pneumonia. Am J Respir Crit Care Med 2002, 65: 867 – 903.
9. COCOROS NM, PRIEBE G, GRAY JE, et al. Factors associated with pediatric ventilator-associated conditions in six U.S. hospitals: A nested case-control study. Pediatr Crit Care Med 2017, 18: e536 – e545.
10. DMYTRIIEV D, DOBROVANOV O, KRALINSKY K, et al. A case report of successful experience of using adaptive support ventilation in the pediatric patient with viral interstitial pneumonia COVID-19 positive. Lek Obz 2021, 70 (3): 119 – 123.
11. DMYTRIIEV D, DOBROVANOV O, KRALINSKY K, BABELA R. Adaptive supportive ventilation in a child with coronavirus pneumonia and diabetes mellitus. Ros Vestn Perinatol Pediat 2020, 65 (5): 66 – 72. DOI: 10.21508/1027-4065-2020-65-5-66-72
12. DMYTRIIEV D, DOBROVANOV O, NAZARCHUK O, et al. Efficacy of inhaled antibiotics in infants with ventilator-associated pneumonia. Lek Obz 2022, 71 (6 – 7): 237 – 240.
13. DMYTRIIEV D, MELNICHENKO M, DOBROVANOV O. et al. Perioperative protective-hemodynamic assessment of ASV usage in pediatric surgical patients. Acute Crit Care 2022, 37 (3): 1 – 8. DOI: 10.4266/acc.2022.00297
14. DMYTRIIEV D, NAZARCHUK O, MELNYCHENKO M, LEVCHENKO B. Optimization of the target strategy of perioperative infusion therapy based on monitoring data of central hemodynamics in order to prevent complications. Frontiers in Medicine 2022, 9, 935331. DOI: 10.3389/fmed.2022.935331
15. DMYTRIIEV K, MOSTOVOY Y, DOBROVANOV O, et al. Efficacy of combination of tiotropium/olodaterol in patients with COPD in real clinical practice. Wiad Lek 2022, 75 (12): 2953 – 2957. DOI: 10.36740/WLek202212110
16. DOBROVANOV O, DMYTRIIEV D, PROCHOTSKY A, et al. Pain in COVID-19: Quis est culpa? Electron J Gen Med 2023, 20 (1): em435. DOI: 10.29333/ejgm/12672
17. DOBROVANOV O, FURKOVÁ K. Pandémia COVID-19: aktuality. Herba: Bratislava 2022, 64 s. ISBN 978-80-8229-023-6.
18. DUDECK MA, WEINER LM, ALLEN-BRIDSON K, et al. National Healthcare Safety Network (NHSN) report, data summary for 2012, Device-associated module. Am J Infect Control 2013, 41 (12): 1148 – 1166.
19. DUFLO F, DEBON R, MONNERET G, et al. Alveolar and serum procalcitonin: diagnostic and prognostic value in ventilator-associated pneumonia. Anesthesiology 2002, 96 (1): 74 – 79.
20. EUROPEAN CENTRE FOR DISEASE PREVENTION AND CONTROL: Surveillance of healthcare-associated infections in Europe 2007. Stock ECDC 2012.
21. FAGON JY, CHASTRE J, WOLFF M, et al. Invasive and noninvasive strategies for management of suspected ventilator-associated pneumonia. A randomized trial. Ann Intern Med 2000, 132 (8): 621 – 630.
22. FOGLIA E, MEIER MD, ELWARD A. Ventilator-associated pneumonia in neonatal and pediatric intensive care unit patients. Clin Microbiol Rev 2007, 20: 409 – 425.
23. GIBOT S, CRAVOISY A, LEVY B, et al. Soluble triggering receptor expressed on myeloid cells and the diagnosis of pneumonia. N Engl J Med 2004, 29, 350 (5): 451 – 458.
24. GIGLIO MT, MARUCCI M, TESTINI M, BRIENZA N. Goal-directed haemodynamic therapy and gastrointestinal complications in major surgery: a metaanalysis of randomized controlled trials. Br J Anaesth. 2009, 103: 637 – 646. DOI: 10.1093/bja/ aep279
25. HORAN TC, ANDRUS M, DUDECK MA. CDC/NHSN surveillance definition of health care-associated infection and criteria for specific types of infections in the acute care setting. Am J Infect Control 2008, 36: 309 – 332.
26. INCHAI J, POTHIRAT C, LIWSRISAKUN C, et al. Ventilator-associated pneumonia: epidemiology and prognostic indicators of 30-day mortality. Jpn J Infect Dis 2015, 68 (3): 181 – 186.

27. ISHIHARA H, SUGO Y, TSUTSUI M, et al. The ability of a new continuous cardiac output monitor to measure trends in cardiac output following implementation of a patient information calibration and an automated exclusion algorithm. J Clin Monit Comput 2012, 26: 465 – 471.
28. JAKRIBETTU R, BOLOOR R. Characterisation of aerobic bacteria isolated from endotracheal aspirate in adult patients suspected ventilator associated pneumonia in a tertiary care center in Mangalore. Saudi Journal of Anaesthesia 2012, 6: 115.
29. JANSSENS S, BEYAERT R. Role of toll-like receptors in pathogen recognition. Clin Microbiol Rev 2003, 16: 637 – 646.
30. KANZLER H, BARRAT FJ, HESSEL EM, COFFMAN RL. Therapeutic targeting of innate immunity with toll-like receptor agonists and antagonists. Nat Med 2007, 13: 552.
31. KLOMPAS M, KLEINMAN K, MURPHY MV. Descriptive epidemiology and attributable morbidity of ventilator-associated events. Infect Control Hosp Epidemiol. 2014, 35: 502 – 510.
32. KOENIG SM, TRUWIT JD. Ventilator-associated pneumonia: diagnosis, treatment and prevention. Clin Microbiol Rev 2006, 19: 637 – 657.
33. KOLLEF MH. Ventilator-associated pneumonia. A multivariate analysis. JAMA 1993, 270: 1965 – 1970.
34. LEWIS SC, LI L, MURPHY MV, et al. CDC Prevention Epicenters: Risk factors for ventilator-associated events: A case-control multivariable analysis. Crit Care Med 2014, 42: 1839 – 1848.
35. LIEW FY, XU D, BRINT EK, O'NEILL LA. Negative regulation of toll-like receptor-mediated immune responses. Nature reviews Immunology 2005, 5 (6): 446 – 458.
36. LIU J, ZHANG S, CHEN J, et al. Risk factors for ventilator-associated events: A prospective cohort study. Am J Infect Control 2019, 47: 744 – 749.
37. MARTIN-LOECHES I, POVOA P, RODRÍGUEZ A, et al. Incidence and prognosis of ventilator-associated tracheobronchitis (TAVeM): a multicentre, prospective, observational study. Lancet Respir Med 2015, 3 (11): 859 – 868.
38. MATHAI AS, PHILLIPS A, ISAAC R. Ventilator-associated pneumonia: a persistent healthcare problem in Indian intensive care units! Lung India 2016, 33: 512 – 516.
39. MEDURI GU, MAULDIN GL, WUNDERINK RG, et al. Causes of fever and pulmonary densities in patients with clinical manifestations of ventilator-associated pneumonia. Chest 1994, 106 (1): 221 – 235.
40. MEREGALLI A, OLIVEIRA RP, FRIEDMAN G. Occult hypoperfusion is associated with increased mortality in hemodynamically stable, high-risk, surgical patients. Crit Care 2004, 8: R60 – 65. DOI: 10.1186/cc2423
41. MORRIS AC, KEFALA K, SIMPSON AJ, et al. Evaluation of the effect of diagnostic methodology on the reported incidence of ventilator-associated pneumonia. Thorax 2009, 64 (6): 516 – 522.
42. NAZARCHUK O, NAGAICHUK V, BAHNIUK N, et al. Susceptibility to Antimicrobials of Acinetobacter baumannii and Pseudomonas aeruginosa Clinical Strains and Their blaVIM Variants in ICU of Regional Burn Centre. Lek Obz 2023, 72 (1): 18 – 23.
43. NAZARCHUK OA, DMYTRIIEV DV, DMYTRIIEV KD, et al. Characteristics of infectious complications in critically ill patients. Wiad Lek 2018, 71 (9): 1784 – 1792.
44. ODDO M, POOLE D, HELBOK R, et al. Fluid therapy in neurointensive care patients: ESICM consensus and clinical practice recommendations. Intensive Care Med 2018, 44: 449 – 463.
45. O'NEILL LA. TLRs: Professor Mechnikov, sit on your hat. Trends in immunology 2004, 25 (12): 687 – 693.
46. PORZECANSKI I, BOWTON DL. Diagnosis and treatment of ventilator-associated pneumonia. Chest 2006, 130: 597 – 604.
47. RELLO J, OLLENDORF DA, OSTER G, et al. Epidemiology and outcomes of ventilator-associated pneumonia in a large US database. Chest 2002, 122 (6): 2115 – 2121.
48. RESTREPO MI, ANZUETO A, ARROLIGA AC, et al. Economic burden of ventilator-associated pneumonia based on total resource utilization. Infect Control Hosp Epidemiol 2010, 31 (5): 509 – 515.
49. RODRÍGUEZ A, PÓVOA P, NSEIR S, et al. Incidence and diagnosis of ventilator-associated tracheobronchitis in the intensive care unit: an international online survey. Crit Care 2014, 18 (1): R32.
50. SAUGEL B, CECCONI M, WAGNER JY, REUTER DA. Noninvasive continuous cardiac output monitoring in peri-operative and intensive care medicine. Br J Anaesth 2015, 114: 562 – 575. DOI: 10.1093/bja/aeu447
51. SAUGEL B, VINCENT JL, WAGNER JY. Personalized hemodynamic management. Curr Opin Crit Care 2017, 23: 334 – 341. DOI: 10.1097/MCC.0000000000000422
52. SCHULTSZ C, BOOTSMA MC, LOAN HT, et al. Effects of infection control measures on acquisition of five antimicrobial drug-resistant microorganisms in a tetanus intensive care unit in Vietnam. Intensive Care Med 2013, 39 (4): 661 – 671.
53. SELIGMAN R, PAPASSOTIRIOU J, MORGENTHALER NG, et al. Copeptin, a novel prognostic biomarker in ventilator-associated pneumonia. Crit Care 2008, 12 (1): R11.
54. SHAHIN J, BIELINSKI M, GUICHON C, et al. Suspected ventilator-associated respiratory infection in severely ill patients: a prospective observational study. Crit Care 2013, 17 (5): R251.
55. SHAPRYNSKYI V, NAZARCHUK O, FAUSTOVA M, et al. Some aspects of infectious complications in patients with surgical diseases. Niektoré aspekty infekčných komplikácií u pacientov s chirurgickými chorobami Multicentrická štúdia. Lek Obz 2020, 69: 257 – 260.
56. SILVERSIDES JA, FITZGERALD E, MANICKAVASAGAM US, et al. Role of Active Deresuscitation After Resuscitation (RADAR) Investigators: Deresuscitation of patients with iatrogenic fluid overload is associated with reduced mortality in critical illness. Crit Care Med 2018, 46: 1600 – 1607.
57. SINGER M, DEUTSCHMAN CS, SEYMOUR CW, et al. The third international consensus definitions for sepsis and septic shock (Sepsis-3). JAMA 2016, 315: 801 – 810.
58. VINCENT JL, SAKR Y, SPRUNG CL, et al. Sepsis in European intensive care units: results of the SOAP study. Crit Care Med 2006, 34 (2): 344 – 353.
59. ZARB P, COIGNARD B, GRISKEVICIENE J, et al. The European Centre for Disease Prevention and Control (ECDC) pilot point prevalence survey of healthcare-associated infections and antimicrobial use. Euro Surveill 2012, 17 (46): 20316.
60. ZIMLICHMAN E, HENDERSON D, TAMIR O, et al. Health care-associated infections: a meta-analysis of costs and financial impact on the US health care system. JAMA Intern Med 2013, 173 (22): 2039 – 2046.

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