HOSPITAL PHARMACOLOGY
International Multidisciplinary Journal

ISSN 2334-9492 (Online)

Hospital Pharmacology. 2019; 6(2):785-793

UDC: 615.33.015.8:579.841.1; 616-022.36:579.841.1

doi:10.5937/hpimj1902785J

Original Study

Pandrug-Resistant Pseudomonas Aeruginosa Isolated from Qualitative Endotracheal Aspirate Could Rather be Contaminant than Causative Agent of Respiratory Infections in Intensive Care Unit Patients: Case Study

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SUMMARY

Introduction: Differentiating colonization from infection is not straightforward, and sometimes antibiotics are prescribed unnecessarily if a clinician relies only on susceptibility report from microbiology lab, further promoting antimicrobial resistance.
Aim: The aim of our study was to investigate whether pandrug-resistant Pseudomonas aeruginosa (PA) isolated from qualitative endotracheal aspirates of ICU patients was primarily external colonization from environmental reservoirs.
Subjects and Methods: An instrumental case study was conducted using qualitative research methodology. An ICU with level 2 or 3 of intensive care from Clinical Center Kragujevac, Serbia, was chosen for the case, and research questions were triangulated by direct observation, interviewing personnel of the ICU and by epidemiological survey.
Results: Pandrug-resistant PA was present in environment of the ICU because hygiene was not stringently kept. It eventually arrived to respiratory circuits of mechanically ventilated patients and gradually descended to endotracheal tube and trachea. Reliance on qualitative endotracheal aspirate in patients with suspected respiratory tract infection led to diagnosing colonization as infection in 50% of cases with isolation of PA. Inadequate hygiene and avoidance of aseptic working techniques together with understaffing and insufficient funding of the ICUs leads to contamination of personnel and environment with Pseudomonas aeruginosa coming from ill patients. It is then cross-transferred to other patients, who are over-treated with reserve antibiotics due to low specificity of qualitative microbiological analyses and the fact that 50% of patients are only colonized.
Conclusions: Pressure made by antimicrobial treatment of colonization creates at first multi drug-, and then pandrug-resistant clones of PA which gradually populate

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environment of the ICU, becoming serious threat for new patients. This vicious spiral could be broken by improvement of hygiene, provision of enough trained personnel and necessary materials, continuous control of personnel's compliance to working standards for ICUs and by introduction of quantitative microbiological analysis of samples.

Keywords: contaminants, drug resistance, bacterial, prescribing, anti-bacterial agents, case study

INTRODUCTION

The growing resistance of bacteria isolated from inpatients to antibiotics is a serious problem all over the world, but in certain regions it reached an alarming level, e.g. in Eastern Europe and Latin America, where 37.7% and 19.1% of isolates were Extended-spectrum β-lactamases (ESBL) – producing Gram–negative microorganisms, respectively [1]. At intensive care units (ICUs) about 4.4% of patients develop nosocomial pneumonia, further 4.4% bloodstream infection and 1% urinary tract infection, mostly associated with intubation, intravenous catheters, and urinary catheters, respectively [2]. Almost a third (28.6%) of all infections in ICUs are caused by antibiotic-resistant organisms [3], which require prescribing of reserve antibiotics. However, differentiating colonization from infection is not straightforward, and sometimes antibiotics are prescribed unnecessarily if a clinician relies only on susceptibility report from microbiology lab [4], further promoting antimicrobial resistance.

In the fall of 2017 new anti-Pseudomonas antimicrobial ceftolozane/tazobactam received marketing authorization in Serbia, and subsequently, preparations were made for introducing this drug in clinical practice at Clinical Center Kragujevac, one of four top-tertiary care hospitals in Serbia. Among the actions undertaken by hospital management in this direction one was to make a survey of the susceptibility of isolates of Pseudomonas aeruginosa to ceftolozane/tazobactam from the patients in central ICU. At the beginning of 2018 the first reports came from the hospital's microbiology lab, all related to Pseudomonas aeruginosa isolated from tracheal aspirates. Both clinicians and hospital management were confused with the results - all isolates (at least a dozen) were pandrug-resistant to all tested antibiotics, including ceftolozane/tazobactam, carbapenems, piperacillin/tazobactam and colistin. Although the reports were based only on isolation of Pseudomonas of aeruginosa (without a threshold), and not on quantitative endotracheal aspirate [5, 6], and ceftolozane/tazobactam was never used previously in this hospital, a doubt on efficacy of this new antibiotic was raised among the clinicians and threatened to hamper its introduction in clinical practice.

In order to resolve this problem, a research group was formed composed of an epidemiologist, clinical pharmacologist and two anesthesiologists/intensive medicine specialists, who formulated the following key questions: (1) is Pseudomonas aeruginosa present in the environment of the ICU due to inadequate hygiene; (2) is Pseudomonas aeruginosa transferred to trachea of intubated patients from the ICU environment; and (3) whether pandrug-resistant Pseudomonas aeruginosa isolated from tracheal aspirates is mostly contaminating instead of causative agent of nosocomial pneumonia, leading to inappropriate prescribing of antibiotics.

AIM

The principal aim of our study was to investigate whether pandrug-resistant Pseudomonas aeruginosa (PA) isolated from qualitative endotracheal aspirates of ICU patients was primarily external colonization from environmental reservoirs.

SUBJECTS AND METHODS

Subjects of this academic non-interventional study were patients and health workers from the ICU with level 2 or 3 of intensive care at Clinical Center Kragujevac, Kragujevac, Serbia. The study was approved by the Eth-

ics Committee of Clinical Center Kragujevac (decision No 01-432011) and consent for conducting the study was obtained from Head of the ICU in Clinical Center Kragujevac, as well as from the study subjects. It was conducted from March the 21st, to April the 24th, 2018, on several occasions which included observation of the ICU's staff behavior and environment, interviews and review of documents.

Methods

Since quantitative research methods cannot offer answers to the research questions implied, an instrumental case study as a qualitative research method was chosen [7]. This non-interventional, observational and empathic study was conducted through the following phases: defining the case (the ICU at Clinical Center Kragujevac), formulating research questions, identification of informers (physicians and nurses from the ICU familiar with the situation in the ICU and willing to collaborate), identification of data sources (patient files, results of epidemiological surveys of environment of the ICU made by an independent institution, Institute for Healthcare, Kragujevac, Serbia), observing behavior of the ICU's staff, interviewing informers from the ICUs staff, direct interpretation of the data, categorical aggregation, methodological triangulation (confirming conclusions by matching results of observation, interviewing and analysis of documents) and writing a report.

Statistical Analyses

This study contains no statistics, being by its nature qualitative. Instead, narratives were used to present the results.

RESULTS

The ICU in Clinical Center Kragujevac is 17-bed unit divided into two 8- and 7-bed rooms connected by ashort corridor to which the doors of two single-bed isolation rooms are opening. However, often one to two additional beds are added to the two large rooms. During the morning shift up to 10 nurses and two to three physicians care for the patients, while during the afternoon and night shifts the staff is reduced to 4-5 nurses and one physician. This is an ICU with level 2 or 3 of intensive care for the entire hospital with 1,200 beds, and admits both surgical and non-surgical patients.

Observation of the ICU's staff behavior by two independent observers revealed that both physicians and nurses are wearing neither protective head caps nor masks when approaching patients (a long hair was noticed by one of the observers attached to one of the syringe pumps, not currently in use). More than half of the female staff members were wearing jewelry on fingers and wrists, as well as artificial nails protruding above tips of the fingers. Physicians and nurses were also leaving the ICU (when called for emergencies) and coming back from general care wards without changing uniforms or using protective gowns or shoe covers. Consultants coming to ICU from other wards (e.g. from Surgery ward) did not put on protective gowns, caps, shoe covers and gloves when entering the ICU and approaching patients. Besides, the ICU personnel washes their uniforms at their homes, without any sterilization process, except ironing.

Some of the nurses were not wearing gloves regularly when caring for patients; often both procedures that should be performed with aseptic technique (like introducing peripheral venous catheter or intravenous blood sampling) and procedures for cleaning patient or changing wound dressings were done without changing gloves in between. It was obvious that at least some of the nurses were wearing gloves for their own protection only, not taking care about the protection of the patients from infection. Frequently, nurses were typing data into ICU computers and caring for patients with the same gloves on hands. After taking off a pair of gloves, only a few nurses washed their hands, while the majority just put on another pair. Dispensers with isopropyl alcohol for hand disinfection were present in both large rooms (with 8 and 7 beds), but they were never used during the two 2-hours observation periods. When asked if they were aware of national Guidelines for hand hygiene in healthcare facilities [8], the majority of nurses said they were, but they could not follow them due to a shortage of time. "I do not know where is my head during morning shift!" – said one of the nurses. From an interview with Head of the ICU we understood that there was recent (a couple of months ago) replacement of the nurses in the ICU (because many experienced nurses left for work abroad

in the EU countries), and more than a half of new ones were young and inexperienced.

The door of the room for collecting contaminated and dirty materials from the patients (including patients' excreta) opens directly to the corridor that connects all four rooms of the ICU. After collection, dirty and contaminated materials are transported through one of the big rooms with patients, coming into close contact with clean materials. When asked if there is some specific time schedule for transporting dirty and contaminated materials, the worker in charge of this job said: "I take it out when the bags are full."

Interviewed nurses (Deputy chief nurse of the department and two other nurses) confirmed that floor and surfaces of furniture and equipment were cleaned five times per day and wiped with diluted alcohol or other commercial disinfectant containing a mixture of ethanol, isopropyl alcohol and polyhexanide. However, they admitted that certain equipment which was borrowed from other hospital wards (like hemodialysis machine) and currently in the ICU were not cleaned by them at all, and they could not recall who cleaned them and when for the last time. Patient beds are also regularly wiped with disinfectants, but never sterilized (although bed clothing is changed at least daily). Ultraviolet lamps for reducing the number of microorganisms in the air exist in each of the rooms, but in the two 6-bed rooms they have not been used recently. "We never turned them on, because these rooms are constantly occupied by patients, and we do not have a safe place to transfer them even for only 12 hours." – said the Deputy Chief nurse.

There were about 8 mechanical ventilators in function, one of them portable. Each patient was connected to a ventilator by single-use tubing with a microbiological filter in front of the endotracheal tube. Deputy Chief nurse said that tubing was never shared between the patients and was disposed of when a patient did not need mechanical ventilation further. However, the portable ventilator was using room air, and there were two filters on its back that had not been recently washed (at least not in the last week), and never sterilized. Other 7 ventilators were connected to the central supply of compressed air; tubing that connects the ventilators and plugs of the central supply were never changed or disinfected. Large compressors that supply the air for the entire hospital are based in closed, subterranean rooms, using air from those rooms that the authors found difficult to breathe in. Air filters used by the compressors are changed every 3 months or when indicators show that it is time for changing. As the workers in charge of the compressors said, no aseptic techniques are used, "we just screw them off and on, and that's it." Oxygen is also supplied centrally, and oxygen dispensers are just plugged in the central supply plugs; however, oxygen dispensers are never disinfected or sterilized, only plastic flask with water is changed for each patient ("but we always change the plastic flask, it can't be missed" – said one of the nurses.

There were also several items in the ICU that are never either cleaned or disinfected. Several such items are computers, keyboards, and computer mouse, which are placed in the two large rooms, and constantly and interchangeably used by nurses, ICU physicians and consulting physicians. Windows on the ICU open towards inside, and since they were never cleaned outside, when open, they shed outside dirt on the floor, including dried excreta of pigeons and other birds. The Deputy Chief nurse said "the problem is that outer surface of the windows could only be cleaned outside, so we cannot do this, but hospital management has to purchase such cleaning service regularly." Finally, some of the long wall lamps above the heads of the patients had a broken glass cover, and inside were thick layers of dust, indicating that no attempts were made to repair the glass for a considerable period of time.

Interviews with two ICU physicians (Head of the ICU and Head of Center for anesthesia and resuscitation) revealed a few additional problems. Visits of the ICU patients by their relatives and friends are allowed once daily, for half of an hour, and visitors are given protective caps, shoe covers, masks, and gowns; however, due to limited hospital budget, protective gowns are not single-use and disposable, but used repeatedly for several visitors on the same day. Such practice gives additional opportunity for the spread of microorganisms from patient to patient. Another important problem is reliance on qualitative tracheal aspirate for the diagnosis of nosocomial pneumonia, which is frequently misleading; this situation was explained by one of the interviewed physicians as "catheters for bronchoalveolar aspiration are not refundable

by the Health Insurance Fund and therefore hospital does not buy them, while on the other hand people from microbiology lab is not acquainted with quantitative tracheal aspirate technique." The physicians also agreed that "some of younger colleagues do not follow the aseptic procedure properly when introducing a central venous catheter, which never happened the past." They also stressed that the ICU is heavily understaffed according to developed countries standards, where a minimum of 2 nurses per bed is a rule.

Epidemiological surveys

Epidemiologist employed in Clinical Center invited microbiology service from other institution (Institute for Healthcare in Kragujevac) to make epidemiological survey of the ICU on two occasions. Samples for microbiological analysis and culture were taken from the following sites in the ICU: room air (n = 6), interior of plastic tubing that connects patients with mechanical ventilators (n = 5), hands of nurses (n = 8), connector for ambu-bag (n = 1), surface of instrument table (n = 2), surface of table for preparing intravenous therapy (n = 3), open bottle of physiological solution for wound irrigation (n = 1), bed clothing (n = 2), direct laryngoscope (n = 1), unused endotracheal tube (n = 1), sterilized surgical instrument (n = 1), sterilized gauze swab (n = 1), solution of povidone iodine (n = 1), liquid soap (n = 1), hand basin (n = 1), wall (n = 1), unused bed cover (n = 1), unused bronchoscope (n = 1), aspirator (n = 2), thoracic drainage machine (n = 1), sterile tube for oxygen supply (n = 1), mechanical ventilator connection for a patient's tubing (n = 2) and bed mattress (n = 1). There were three isolates of pathogenic bacteria, and in all cases it was pandrug-resistant Pseudomonas aeruginosa; it was isolated from bed clothing (n = 1), open bottle of physiological solution for wound irrigation (n = 1) and interior of plastic tubing that connected a patient with mechanical ventilator (n = 1).

Answers to key research questions

Pseudomonas aeruginosa was obviously present in the environment of the ICU, as shown by the epidemiological survey. There are many reasons to blame inadequate hygiene for its presence: non-adherence to hand washing guidelines, avoidance of protective clothing by the ICU staff, inadequate use of gloves, crossings of clean and dirty material pathways, transfer of microorganisms from patient to patient on gowns worn by visitors, using keyboards with gloves previously used with patients, wearing hand and wrist jewelry, rare use of UV lamps for decontamination of air and surfaces and some never-cleaned spots as lamps and windows from outside. Although some of the hygiene breaches could be excused by understaffing and limited hospital budget, the majority are correctable only by full implementation of appropriate procedures and control of adherence to the procedures.

Answer to the second question is somewhat more difficult to give, as behavior that would connect directly Pseudomonas from the environment with trachea of the ICU patients was neither observed nor implicated from the interviews. However, pandrug-resistant Pseudomonas aeruginosa was isolated from tubing connecting mechanical ventilator with an endotracheal tube of a patient, where it could come from the environment. Another thing is complete microbiological unsafety of air that is pumped into the lungs of mechanically ventilated patients; the only barrier to eventual pathogenic microorganisms is a bacterial filter in front of the endotracheal tube. Whether microorganisms may pass this barrier and under what circumstances remains to be elucidated.

The ICU physicians were aware that qualitative tracheal aspirate may lead to false positive diagnosis of ventilator-associated pneumonia, although clinical outcomes are similar as when quantitative cultures or more invasive sampling methods were used [9]. However, lack of funding and weak interest of microbiologists to offer quantification of endotracheal samples led to reliance only on qualitative endotracheal aspirates in the observed ICU, which bring to the light significant percentage of pandrug-resistant contaminants from the ICU environment instead of true causative agents. Although individual patient may not lose too much if treated for contamination with reserve antibiotics, in the long run this creates pressure for further selection of pandrug-resistant strains of many bacteria including Pseudomonas, and may preclude timely use of antibiotics that were falsely judged as inappropriate according to resistance pattern of isolated contaminate.

DISCUSSION

Our study showed that pandrug-resistant Pseudomonas aeruginosa was present in the environment of the ICU because hygiene was not stringently and continuously kept. It eventually arrived to respiratory circuits of mechanically ventilated patients and gradually descended to endotracheal tubes and trachea. If the only qualitative endotracheal aspirate is used for microbiological diagnosis of suspected respiratory tract infection (it is usually ventilator-associated pneumonia that is suspected), the contaminate could be misunderstood as a causative agent. Being pandrug-resistant due to continuous exposure to antimicrobial agents in the ICU environment, such an isolate of Pseudomonas aeruginosa will provoke prescribing of reserve antibiotics, often in high doses and combinations, further promoting antimicrobial resistance.

It was shown in a recent study of Harris et al [10] that 11.6% of patients are colonized with Pseudomonas aeruginosa on admission to an ICU. Colonization bears risk not only for those who are colonized (28.2% of them become culture positive for Pseudomonas aeruginosa during the same hospitalization), but also for other patients in the ICU, as Pseudomonas aeruginosa quickly spreads to the environment. It was isolated in our study from bed clothing, physiological solution and respiratory tubing, and in a study of Boyer et al even 31% of environmental samples from an ICU were positive for Pseudomonas aeruginosa [11]. Transfer of Pseudomonas in opposite direction, from the environment to the patients, is also intensive, as in the same study 16% of patients who were not colonized at admission acquired Pseudomonas aeruginosa during their stay in the ICU.

Cross-transmission of Pseudomonas aeruginosa from the environment and personnel of an ICU to patients is responsible for 59.5% of either infection or colonization cases [12], and it could be prevented with stringent hygiene and aseptic techniques when inserting catheters and other devices to the patients. Exogenously acquired Pseudomonas aeruginosa is predominantly multidrug-resistant, and originates from one or just a few clones, like in the study of Agodi et al where 60.8% of isolates from patients belonged to the same clone [12]. Pseudomonas aeruginosa isolates in our study, both from patients and the environment, were pandrug-resistant, but we were not technologically equipped enough to investigate their clonal origin. Cross-transmission of multidrug-resistant Pseudomonas aeruginosa from patient to patient goes through ICU personnel and environment, resulting in either colonization or infection (the approximate ratio is 50% : 50%) [13]. Although treating infections caused by multidrug-resistant Pseudomonas aeruginosa is a difficult task, administration of second- and third-line antibiotics to patients who are only contaminated creates additional problems, inducing selection of pandrug-resistant strains which are then again involved in the cross-transmission circle. Since all isolates in the ICU that we investigated, were pandrug-resistant to antimicrobials, it seems that this ICU already came to the bottom of the spiral, after passing numerous circles.

Colonization of tubing and respiratory tract of the ICU patients with Pseudomonas aeruginosa which originates from environmental reservoirs was demonstrated previously [14]. Patients on mechanical ventilation more frequently become carriers of P. aeruginosa during their stay in the ICU than other patients [15]. Although it was shown that the tap water was the major reservoir of Pseudomonas aeruginosa in ICUs, which is responsible for exogenous colonization of the patients [16], other sources could also be significant, such as the air which is pumped into the patients' lungs by mechanical ventilators. In our study mechanical ventilators were never sterilized by ethylene oxide, and only wiped with disinfectants once daily; besides, quality of air insufflated into patients' lungs was never checked. Indeed, study by Sui et al identified Pseudomonas aeruginosa on the surfaces of Y-branching (6.7%) and water traps (13.3%) of breathing circuits, and showed that mechanical ventilators and their tubing had to be disinfected as often as every 8 hours in order to avoid colonization of the patient's respiratory tract [17]. Endotracheal colonization with pandrug-resistant Pseudomonas aeruginosa in our study was suggested by all three research methods applied: observation of breaches of hygienic principles, information about maintenance of mechanical ventilators gathered from interviews with personnel and epidemiological survey.

The main limitation of our study is

the lack of genotyping of Pseudomonas aeruginosa isolates, as this would help us explain the origin and pathways of circulation of this microorganism in the ICU. Besides, not all relevant samples from the ICU environment were taken for culturing, especially tap water, which was already underlined in previous studies as one of the most important environmental reservoirs.

Since the authors conducted several visits to the ICU to make repeated observations and interviews, an interesting behavior took place during the course of the study. After the staff understood from the investigators' questions and their interest in certain details what was wrong concerning hygiene, they corrected their practice and cleaned certain items more thoroughly. Already after the first visit made by investigators, lamps above the patients, that had never been cleaned before, were completely clean and disinfected, and their broken protective glass replaced with a new one. It was also noted that air filters on a mobile mechanical ventilator had been washed and disinfected, nurses stopped wearing hand and wrist jewelry, and artificial nails, the practice of handwashing became compliant with national guidelines, and personnel started to wear gloves according to the recommendations.

CONCLUSION

In conclusion, inadequate hygiene and avoidance of aseptic working techniques together with understaffing and insufficient funding of the ICUs leads to contamination of both staff and the environment with Pseudomonas aeruginosa coming from patients. It is then cross-transferred to other patients, who are over-treated with reserve antibiotics due to low specificity of qualitative microbiological analyses and the fact that 50% of patients are only colonized. The pressure made by antimicrobials creates at first multi drug-, and then pandrug-resistant clones which gradually populate the environment of the ICU, becoming a serious threat for new patients. This vicious spiral could be broken by an improvement of hygiene, provision of enough trained personnel and necessary materials, continuous control of personnel's compliance to working standards for ICUs and by the introduction of quantitative microbiological analysis of samples.

CONFLICT OF INTEREST

None.

FINANCIAL DISCLOSURE STATEMENT

The authors were not paid for this work by any interested party.

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Panrezistentan pseudomonas aeruginosa izolovan iz kvalitativnog trahealnog aspirata je verovatnije kontaminat nego uzročnik respiratornih infekcija kod pacijenata u intenzivnoj nezi: studija slučaja

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KRATAK SADRŽAJ

Uvod: Razlikovati kolonizaciju od infekcije nije uvek lako u klničkoj praksi, i ponekada se antibiotic nepotrebno propisuju ako se kliničar oslanja samo na rezultate antibiograma, što dalje promoviše rezistenciju mikroorganizama na antibiotike.
Cilj: Cilj naše studije je bio da se ispita da li panrezistentan Pseudomonas aeruginosa (PA), izolovan iz kvalitativnog trahealnog aspirata pacijenata u intenzivnoj nezi, predstavlja primarno kolonizaciju iz spoljašnjih rezervoara ove bakterije.
Metodologija: Sprovedena je instrumentalna studija slučaja uz korišćenje kvalitativnih metoda istraživanja. Jedinica intenzivne nege sa nivooom intenzivnog lečenja 2 ili 3 Kliničkog centra Kragujevac je bila izabrana za slučaj, dok su odgovori na istraživačko pitanje traženi metodom triangulacije, koja je uključila direktnu opservaciju, intervjue sa medicinskim osobljem i epidemiološku kontrolu prisustva PA.
Rezultati: Panrezistentni PA je bio prisutan u intenzivnoj nezi pre svega zbog propusta u održavanju higijene. Iz okruženja pacijenata PA je dospevao u plastična creva kojima su pacijenti bili povezani sa aparatima za mehaničku ventilaciju, da bi se postepeno spuštao u endotrahealni tubus i traheu. Oslanjanje na kvalitativan endotrahealni aspirat kod pacijenata sa sumnjom na infekciju respiratornog trakta je dovelo do pogrešne kvalifikacije kontaminacije kao infekcije u oko 50% slučajeva izolacije PA. Neodgovarajuće higijena i izbegavanje korišćenja aseptičnih tehnika rada, zajedno sa nedovoljno osoblja i nedovoljnim finansiranjem jedinica intenzivne nege dovodi do kontaminacije personala i okruženja sa Pseudomonas aeruginosa koji potiče od pacijenata sa infekcijom. Ova bakterija se zatim prenosi na druge pacijente, koji su potom preterano tretirani rezervnim antibioticima zbog niske specifičnosti kvalitativnih mikrobioloških analiza i činjenice da je oko 50% pacijenata sa izolatima ustvari samo kolonizovano.
Zaključak: Pritisak usled sistemske primene antibiotika kod kolonizacije stvara prvo multirezistentne, a zatim i panrezistentne sojeve PA, koji postepeno naseljavaju prostor u intenzivnoj nezi, postajući sve veća opasnost za nove pacijente. Ovaj circulus vitiosus se može prekinuti poboljšanjem higijene, obezbeđenjem dovoljno obučenog osoblja i neophodnih materijala za rad, stalnom kontrolom da li se osoblje pridržava standarda rada u intenzivnoj nezi i uvođenjem kvantitativne mikrobiološke analize trahealnih aspirata.

Ključne reči: kontaminat, rezistencija bakterija na antibiotike, propisivanje lekova, antibakterijski lekovi, studija slučaja

Received: May 13, 2019
Accepted: May 29, 2019