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Automated phenotypic method versus manual method in the identification of microorganisms isolated from blood cultures: clinical and microbiological outcomes

Método fenotípico automatizado versus método manual na identificação de micro-organismos isolados de hemoculturas: desfechos clínicos e microbiológicos

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ABSTRACT

Introduction: Automation is increasingly used in microbiology laboratory, however, few studies assessed clinical outcomes compared to traditional methods. In Brazil, no studies with this objective were detected. **Objective:** To analyze the clinical and microbiological impacts after implantation of an automated phenotypic method in a microbiology service. **Methods:** Observational and retrospective study carried out on the microbiology laboratory involving blood culture test from intensive care unit (ICU) patients. Data were collected from hospitalized patients between January 2014 and December 2015. The length of hospitalization, number of empirical therapies, deaths and information related to microbiological isolation were analyzed. The sample was obtained by convenience. Pearson's Chi-square and Student's t-tests were used to compare outcomes. The program used was the Stata release, version 11, being considered significant values of $p<0.05$. **Results:** A total of 472 patients were evaluated. There was no reduction in the empirical prescription of antimicrobials (54.7% vs 45.3%; p=0.33), ICU stay (14.5 days vs 15.8 days; p=0.78) and mortality (54.4% vs 45.6%; p=0.36). Similarly, profile of isolated agents in both methods did not appear to be discrepant, however, there was an increase of 44.7% in the number of microbial isolates (76 vs 110) and a better characterization of them. **Conclusion:** The microbiology laboratory automation did not modify the length of stay, ICU mortality and the number of empirical therapies. However, identification and isolation of microorganisms was improved.

Keywords: automation; blood culture; microbiology; microbial sensitivity tests.

RESUMO

Introdução: A automação laboratorial é cada vez mais utilizada em microbiologia, no entanto, poucos estudos avaliam desfechos clínicos em comparação aos métodos tradicionais. No Brasil, nenhum estudo com esse objetivo foi detectado. **Objetivo:** Analisar os impactos clínicos e microbiológicos após implantação de método fenotípico automatizado em um serviço de microbiologia. **Métodos:** Realizamos estudo observacional e retrospectivo no laboratório de microbiologia referente a exame de hemocultura de pacientes da Unidade de Terapia Intensiva (UTI). Os dados foram coletados de pacientes internados entre janeiro/2014 a dezembro/2015. Analisou-se o tempo de internação, número de terapias empíricas, óbitos e dados relacionados ao isolamento microbiológico. A amostra foi obtida por conveniência. Para a comparação entre os desfechos foram empregados os testes t de Student e Qui-quadrado de Pearson. O programa empregado foi o Stata release, versão 11, sendo considerados significativos valores de $p<0,05$. **Resultados:** Foram avaliados 472 pacientes. Não houve redução na prescrição empírica de antimicrobianos (54,7% vs 45,3%; p=0,33), tempo de internação na UTI (14,5 dias vs 15,8 dias p=0,78) e na taxa de óbitos (54,4% vs 45,6%; p=0,36). Similarmente, o perfil de agentes isolados em ambos os métodos não parece ser discrepante, no entanto, houve um aumento de 44,7% no número de isolados microbianos (76 vs 110) com melhor caracterização dos mesmos. **Conclusão:** A automação do laboratório de microbiologia não impactou no tempo de internação, mortalidade na UTI e no número de terapias empíricas. No entanto, a identificação e o isolamento de microrganismos melhoraram.

Palavras-chave: automação; hemocultura; microbiologia; testes de sensibilidade microbiana.

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INTRODUCTION

Automation stands out in microbiology laboratories, characterized by gradual changes in laboratory routine with replacement of manual techniques by automated devices, covering several stages of the microbiological examination.

Microbiological automation minimizes errors[1] improves quality of microbiological examination[2] reduces incubation time[3,4] and decreases hospital costs[5]. There are also studies demonstrating the advantages in cost-effectiveness of automated methods based on molecular analysis opposite to the conventional method[6]. However, as disadvantages, the reduced participation of the microbiologist in some preanalytical and analytical stages of the process is highlighted[7]. Automation in microbiology presents high maintenance costs, as well as contamination risks in sample and incubator processors[8].

Among the methods used in the automation of microbiology laboratories, phenotype is widely used. It consists of the detection of colorimetric and turbidity changes in the incubated substrates, these alterations reflect the microbial metabolism allowing identification of pathogen and performance of antibiogram. Examples of systems commercialized is the VITEK® 2, described in literature as agile and accurate for microbiological identification and antibiogram[9-11].

Despite the advantages related to automated microbiology, few studies have evaluated clinical outcomes[8]. As far as we know, some scholars compared the impact of automation implantation on patients and observed shorter hospitalization time, costs reduction, better therapeutic optimization and reduction in mortality[5,12,13]. Therefore, despite the importance of the topic and considering the cost of automation, data are scarce to assist in decision making for its implementation in the services. In Brazil, no studies with this objective were detected.

Therefore, the objective of this study was to evaluate the clinical and microbiological impacts after implantation of an automated phenotypic method (VITEK® 2 compact) in the microbiological identification of blood culture of critical patients.

METHODS

Study design and population

Observational and retrospective study in a microbiology laboratory of a university hospital. Data were collected from patients hospitalized at the Intensive Care Unit (ICU) of the Onofre Lopes University Hospital (HUOL), a reference institution in emergency cardiology and renal transplantation in state of Rio Grande do Norte (a total of 248 beds, 19 in ICU).

Information on critical patients was included with age 18 years and with a blood culture request were included. Patients with incomplete data in electronic medical record and readmitted to ICU were excluded, the latter being considered the data only the first admission. We considered as critical patient all those who needed intensive care in the ICU. For better understanding of the article, the terms VITEK® 2 compact and phenotypic method/automated phenotypic method will be used as synonyms.

This study was approved to Research Ethics Committee of HUOL (CAAE no. 60372116.0.0000.5292).

Data collection

The collection period was between January 2014 and December 2015. The following information was collected: age in years, sex, use of invasive devices and stay in days of the peripheral and central venous accesses on the day of collection of the first blood culture, diagnosis of sepsis and blood culture results. For the clinical outcomes were collected time of intensive care, number of empirical antimicrobial prescriptions (for patients with positive blood culture) and mortality.

The information was obtained from the electronic medical record and the records of the microbiology service.

From January 2014 to February 2015, the processing of samples relating to microbiological identification were performed by manual methods, being called the manual group (Gm). The automated group (Ga) included data from March to December 2015, when the microbiological identification were automated through VITEK® 2 Compact (BioMérieux, France).

Clinical isolates

For both groups, Gm and Ga, clinical isolates were obtained from positive blood previously incubated in BacT/Alert® 3D (BioMérieux, France) automated system at 36.5°C, system that detects only positive samples. Positive vials were submitted to bacterioscopy (Gram staining) and the sample was seeded in the culture media in blood, McConkey and mannitol agar (incubated at 36.5ºC for 24 hours) for identification of the micro-organism and subsequent performance of the sensitivity test by manual or automated method. In the manual methodology, biochemical tests were used to identify microorganisms: for Gram-negative fermentation tests glucose and lactose, motility, gas production, sulfide, indole, phenylalanine, lysine and ornithine decarboxylase, citrate, malonate and oxidase were perfomed, while for Gram-positive they were made the catalase test, coagulase tube, optochin, bacitracin, esculin bile and PYR (Pyrrolidonil arylamidase). These tests were supplemented by diffusion disc sensitivity tests on specific antimicrobials performed according to Clinical & Laboratory Standards Institute (CLSI) standardization. In the automated methodology the sample was directed to VITEK® 2 Compact: colonies isolated from the culture media were selected and collected with swab and dissolved in a tube containing 3 mL of saline until complete homogenization of the suspension. The turbidity of the suspension was performed by

densichek (Biomérieux, France) respecting the range 0.5 to 0.63 McFarland scale and taken to the apparatus. The microorganisms were identified phenotypically in genus and species by the automated method. Phenotypic identification by the manual method was only possible for some microorganisms, since this method often does not allow a better characterization of some species.

Statistical analysis

The sample was obtained by convenience, searching for periods and number of equivalent samples between the two groups. Statistical analysis was performed using Stata release 11 (Stata Corporation, College Station, TX, USA). In the descriptive analysis, the characteristics of the patients and isolated microorganisms were presented in mean and standard deviation or relative or absolute frequencies when appropriate. For the comparison between the Gm and Ga groups we used the Pearson's Chi-square for the proportions and Student's t-test for the means between the groups. In the case of nonparametric distribution identified by the Shapiro-Wilk test, we used the Mann-Whitney U test for comparison of means. Values of $p<0.05$ were considered significant.

RESULTS

During the study period, 492 patients had a blood culture test, but only 472 met the inclusion criteria (Table 1). Of these, 248 are from the manual group (Gm) and 224 from the automated group (Ga). The mean age was 59.0±17.3 years with a low predominance of females (53.4%). The main invasive devices were the peripheral (58.6%) and central (51.4%) venous accesses, and mean length of stay until blood collection was 3.1±3.6 days. Sepsis was diagnosed in 44.1% of the patients and 82.2% of the cases were those with positive blood cultures (30.9%). Of these, 72.6% had empirically prescribed antimicrobial. The patients had an average of 15.3±16.2 days under intensive care and overall mortality was 45.1%.

In Table 1, population characteristics between Gm and Ga groups are described. Use of a hemodialysis catheter (36.1% vs 63.9%, $p<0.01$), device for invasive blood pressure assessment (32.4% vs 67.7%, $p=0.01$) and greater venous access time (2.7±3.0 vs 3.6±4.2 days, $p<0.01$) was more pronounced in Ga.

There was no reduction in the empirical prescription of antimicrobials ($p=0.33$), ICU stay ($p=0.37$) and mortality ($p=0.36$). As for empirical prescription, there were no significant changes in treatments usually place after automation (Figure 1).

The profile of isolated agents in both methods did not appear to be discrepant, however, there was an increase in the number of microbial isolates and a better characterization of them, with a percentage reduction of agents identified only by genus and identification of species that were not detected (Table 2). By correlating the most commonly prescribed antimicrobials empirically to the microorganisms that were later isolated more frequently, we observed that the Gram-negative species had a greater resistance profile to the treatments usually instituted and the fungal infections were little considered when prescribing the empirical therapy (Table 3), with only 10% of the empirical prescriptions including fluconazole empirically.

DISCUSSION

We observed that the automated phenotypic method, despite a larger number of isolates and better microbiological characterization, did not alter the number empirical prescriptions, as well as time of hospitalization, mortality and to the empirical treatments usually instituted.

The automation of microbiological analysis has improved several aspects in laboratorial work and health care. Taking this into account, the literature reports better sensitivity, shorter incubation time[14] higher reproducibility and quality in the isolation of microorganisms leading to results with lower variability and higher rate of detection in biological samples[2]. Other benefits of automation would be better tracking quality[1], however greater agility in microbiological identification is highlighted[7,15,16]. As for clinical outcomes, automation would reduce errors in medical conduct, mortality, hospitalization time and costs[1,5,8,12,16]. However, among the outstanding advantages, we observed some discrepancies with our data.

Table 1: Clinical and infectious profile and clinical outcomes among patients submitted to manual (Gm) and automated (Ga) method (n=472).

Characteristics	Total (n=472)		Gm (n = 248)		Ga (n = 224)		p*
Age (m, sd)	59.0	17.3	58.5	17.8	59.6	16.6	0.48
Women (n, %)	252	53.4	144	57.1	108	42.9	0.09
Invasive devices (n, %)							
Peripheral venous access	276	58.6	150	54.4	126	45.7	0.38
Central venous access	242	51.4	120	49.6	122	50.4	0.17
Hemodialysis catheter	72	15.3	26	36.1	46	63.9	<0.01
Drains	62	13.2	33	53.3	29	46.8	0.92
Invasive blood pressure	34	7.2	11	32.4	23	67.7	0.01
Permanence of venous accesses (m, sd)	3.1	3.6	2.7	3.0	3.6	4.2	<0.01
Sepsis diagnosis (n, %)	208	44.1	112	53.8	96	46.2	0.62
Patients with positive blood culture (n, %)	146	30.9	75	51.4	71	48.6	0.73
Stay in the ICU on days (m, sd)	15.3	16.2	14.5	13.2	15.8	18.9	0.37
Empirical prescription of ATM (n, %)	106	72.6	58	54.7	48	45.3	0.33
Deaths (n, %)	213	45.1	141	54.4	118	45.6	0.36

n – number of samples; m – mean; sd – standard deviation; **Pearson's chi-squared test* or Student's t-test. ATM – Antimicrobial. *The empirical prescription of ATM was evaluated only for patients with positive blood culture.*

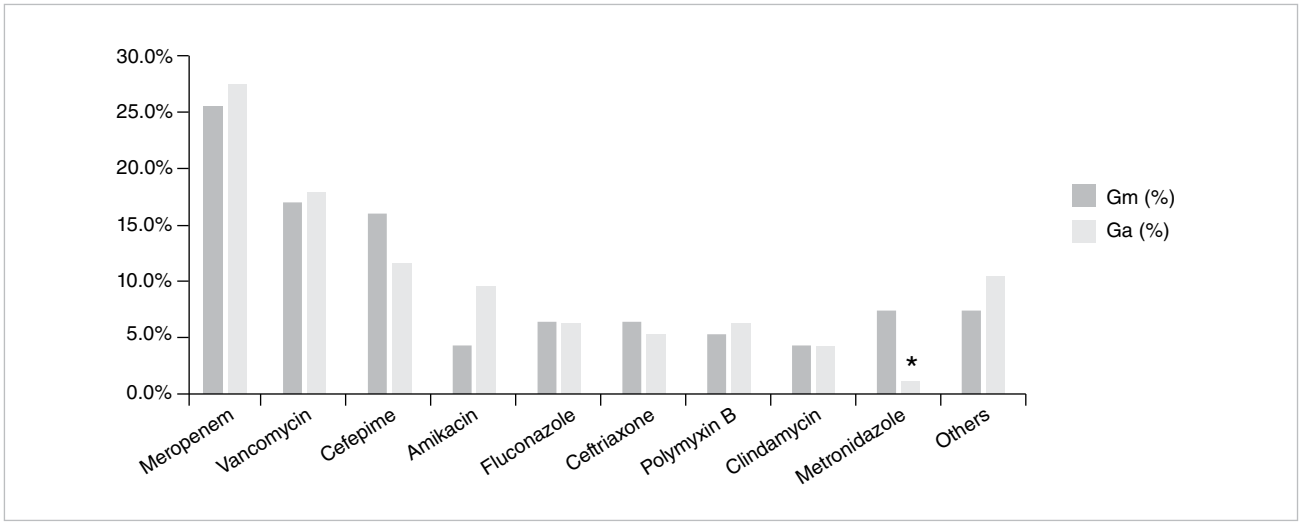

Figure 1: Empirical prescription of antimicrobial. * p<0.05 (Pearson's chi-squared test).

Table 2: Agents isolated by manual method (Gm) and automated method (Ga).

Etiological agent	Gm (n, %)		Ga (n, %)		p*
Acinetobacter spp.	10	13.1	4	3.7	0.03
Acinetobacter baumannii[(a)]	-	-	12	10.9	-
Acinetobacter lwoffii[(a)]	-	-	1	0.9	-
Enterobacter spp.	6	7.9	2	1.8	0.06
Enterobacter cloacae[(a)]	-	-	2	1.8	-
Enterococcus spp.	4	5.3	1	0.9	0.08
Enterococcus faecalis[(a)]	-	-	4	3.7	-
Klebsiella spp.	6	7.9	3	2.7	0.13
Klebsiella pneumoniae[(a)]	-	-	3	2.7	-
Yeasts	4	5.3	6	5.5	0.96
Candida albicans[(a)]	-	-	3	2.7	-
Candida glabrata[(a)]	-	-	1	0.9	-
Candida spp.[(a)]	-	-	1	0.9	-
Candida tropicalis[(a)]	-	-	1	0.9	-
Pseudomonas spp.	5	6.6	4	3.7	0.38
Pseudomonas aeruginosa[(a)]	-	-	3	2.7	-
Coagulase-negative *staphylococci*	20	26.3	26	23.6	0.75
Staphylococcus capitis[(a)]	-	-	2	1.8	-
Staphylococcus epidermidis[(a)]	-	-	4	3.7	-
Staphylococcus haemolyticus[(a)]	-	-	1	0.9	-
Staphylococcus hominis[(a)]	-	-	1	0.9	-
Others	21[(b)]	27.6	13[(c)]	11.8	-
Unidentified agents in Gm[(d)]	-	-	12	10.9	-
Total	76	100	110	100	

n – number of samples; **Pearson's chi-squared test.*
(a) Agents that started to have their respective species identified only after automation. **(b)** *Staphylococcus aureus* (8-10.5%), *Escherichia coli* (5-6.6%), *Proteus vulgaris* (3-4.0%), *Streptococcus spp.* (2-2.6%), *Streptococcus pneumoniae* (1-1.3%), *Proteus spp.* (1-1.3%), *non-fermenting Gram-negative bacilli* (1-1.3%). **(c)** *Staphylococcus aureus* (6 - 5.45%), *Escherichia coli* (4-3.63%), *non-fermenting Gram-negative bacilli* (3-2.72%). **(d)** *Serratia marcescens* (4-3.6%); *Burkholderia cepacia* (2-1.8%), *Achromobacter dentrificans* (1-0.9%), *Aeromonas hydrophila/caviae* (1-0.9%), *Citrobacter freundii* (1-0.9%), *Morganella morgannii* (1-0.9%), *Raoultella ornithinolytica* (1-0.9%), *Stenotrophomonas maltophilia* (1-0.9%).

With a similar design to our study, Huang *et al.*[12] evaluated clinical outcomes in 501 patients with positive blood cultures. The authors observed a shorter hospitalization time, mortality and better optimization of antimicrobial use after submitting the microbiological identification to the automated method. Similarly, Perez *et al.*[5] included 219 patients with positive blood cultures for aerobic Gram-negative or anaerobic facultative, highlighting the therapeutic optimization, decrease in hospitalization time and cost. Other author, Delport *et al.*[13] analyzed data from 396 patients with positive blood cultures and compared the period of the conventional method execution of microbiological identification with the period after automation of this process associated to the creation of a fast identification protocol, obtaining as results the reduction in the microbiological examination runtime, faster therapeutic appropriateness, shorter hospital stay and lower risk of mortality.

The cited articles present some methodological differences compared to our study. The authors investigated hospitalized patients with positive blood cultures. In contrast, we analyzed positive and negative blood cultures. Negative results in microbiological evaluation also guide clinical behavior and impact on costs. Our data encompass the process of automation in all critical patients, allowing a more complete analysis. Another difference was the direct detection of pathogens in clinical samples.

However, an important difference observed is the automated microbiological identification method used by Huang *et al.*[12], Perez *et al.*[5] and Delport *et al.*[13]: *matrix assisted laser desorption ionization-time of flight* (MALDI-TOF). This methodology is technically superior to the VITEK® 2 compact[17,18]. MALDI-TOF identifies microorganisms in minutes, while the automated phenotypic method needs hours for identification. This greater velocity would lead to better clinical results[19,20]. Therefore, the benefits of automation in microbiology are closely related to the type of method. Our study compares

the automated phenotypic method with the traditional method in the identification of microorganisms in blood cultures, we did not detect difference in mortality and hospitalization time.

After automation, however, we observed a better characterization of the microbiological profile (Table 3). Automation improves the quality of the process and increases the recovery of microorganisms in biological samples[2]. In the manual method, the identification was restricted to the genus (for example, *Acinetobacter spp., Enterobacter spp. and Enterococcus spp*). The identification of fungi is more limited, characterized only as yeast. The automated phenotypic method allowed for better microbiological identification. For example, samples of *Acinetobacter spp.* have been identified as *A. baumannii* and *A. lowffii*. Likewise, species of fungi (*Candida albicans, Candida glabrata* and *Candida tropicalis*) became isolated. It was also possible to observe an increase in the number of bacteria and fungi isolated after automation and species that were not detected were identified after automation. Regarding the variability of microorganisms isolated in our study, it does not appear to be very different from other studies, differing only in the incidence of the different agents in the services[3,12,19].

A better characterization of microorganisms influences the selection of antimicrobial therapy. For example, different species of *Candida spp* have different sensitivity to treatment with echinocandins[21] as well as species of the genus *Enterococcus spp.* and its response profile to variable vancomycin[22]. In particular, in critically ill patients, appropriate choice of antimicrobials results in better clinical outcomes[23,24] and less occurrence of microbial resistance.

Several factors are related to prescription of antimicrobials: isolated species, bacterial sensitivity, infection topography, pharmacokinetic aspects and intrinsic characteristics of the patient, among others. Regarding the factors related to empirical prescription, it is possible to emphasize the clinical severity of the patient, in which case the least time to start is primordial, and the knowledge of the infectious profile of the institution. The latter is essential. In our study, we observed that fungal infections have not gained the same importance in the diagnostic hypotheses, although they were a major cause of bloodstream infection. In this group, only 10% of the prescriptions included fluconazole empirically and the mortality rate exceeded 80%. Other antimicrobials used empirically when the isolate was yeast or *Candida spp*. where: meropenem and vancomycin (25% each), cefepime (15%), polymyxin B (10%) and amphotericin B, metronidazole and piperacillin-tazobactam (5% each). As for the sensitivity profile, Gram-negative infections were more resistant to empirical treatments against Gram-positive infections, another factor that impacts the quality of care and the choice of initial empirical treatment.

Adjustments in antimicrobial therapy after initial empirical therapy may result in better outcomes[25,26]. Huang *et al.*[12], Perez *et al.*[5] and Delport *et al.*[13] observed that reduction in time to identify microorganism resulted in rapid adjustments in antimicrobial treatment, however, as highlighted, the

Table 3: Description of the most incident microorganisms, main antimicrobials prescribed empirically by isolated agent and sensitivity profile.

Isolated agent	ATM	Sensitivity		p*
		Gm (n; %)	Ga (n; %)	
Acinetobacter spp.	Amikacin	12; 50.0	21; 71.4	0.55
	Cefepime	12; 25.0	21; 14.3	0.16
	Meropenem	12; 25.0	20; 15.0	0.20
Leveduras/Candida spp.	Fluconazole	**	6; 100.0	-
Staphylococcus aureus	Clindamycin	9; 55.5	9; 44.4	0.64
	Meropenem	*NT*	*NT*	-
	Vancomycin	9; 100.0	9; 100.0	>0.99
Klebsiella spp.	Meropenem	6; 50.0	6; 80.0	0.58
	Polimixina B	6; 100.0	6; 100.0	>0.99
Pseudomonas spp.	Amikacin	6; 66.6	7; 85.7	0.71
	Cefepime	6; 66.6	7; 57.1	0.85
	Meropenem	6; 66.6	8; 50.0	0.67
Enterobacter spp.	Cefepime	5; 80.0	4; 25.0	0.14
	Ceftriaxone	6; 83.3	4; 25.0	0.11
	Meropenem	6; 100.0	3; 100.0	>0.99
Enterococcus spp.	Amikacin	6; 83.3	*NT*	-
	Meropenem	6; 100.0	*NT*	-
	Vancomycin	4; 100.0	6; 100.0	>0.99

NT – Not Tested; n – number of samples tested; *Pearson's chi-squared test.
**Sensitivity tests for fungal isolates were only performed after automation.
Staphylococcus spp. negative coagulase represented 29.03% of the total, but were not included in this table because it represented contamination in more than 90% of the samples. The remaining 17.73% represent the other isolated microorganisms that were not included due to the low number of clinical isolates (Table 2).

MALDI-TOF methodology allows more agile analyzes against the automated phenotypic method. Therefore, despite the shortest execution time in the microbiological identification provided by VITEK® 2 compact against the manual method, it would not be able to provide the agility necessary for adjustments in microbial treatment as fast as the MALDI-TOF, or, in our case, to reduce the number of empirical treatments.

Regarding this type of treatment, an important factor to consider is diagnosis of sepsis prevalent in our sample. The guidelines indicate the initiation of broad-spectrum empirical therapy within a maximum of one hour after diagnosis of sepsis[27]. Considering what was said in the previous paragraph, this variable directly impacts on the number of empirical antimicrobial prescriptions in our study.

This work has some limitations. Data were collected in a single unit from a single institution, reducing the generalizability of the findings. Retrospective collection may be associated with incomplete or incorrect information. We also did not assess whether antimicrobial therapies were adequate after release of microbiological test results, as other studies have done, and therefore we cannot assess whether this factor could be related to a better clinical outcome in our analysis. Finally, we did not evaluate the time of execution of microbiological examination before and after automation.

The differences observed between the groups regarding the use of a hemodialysis catheter, device for assessing invasive blood pressure and access time, were changes related to the hospital service and not to the automation process, that is, did not affect the study results.

As for our methodology, we highlight the analysis of equivalent populations before and after automation, in addition to a considerable period of data collection and number of participants, although with few isolated agents (186 microorganisms).

It is important to note that automation in the microbiology laboratory has multiple benefits. Although we have not observed advantages in relation to clinical outcomes, the literature establishes automation as related to optimization of workflow and in the clinical practice we have observed advantages already highlighted in the literature, such as more agile exams in relation to the manual method, which demonstrates its importance to the service and the continuous need for investments.

In conclusion, the automation of the microbiology laboratory did not bring about significant changes in the empirical treatments usually instituted after automation, did not reduce the number of empirical treatments, length of hospital stay and ICU mortality. However, identification and isolation of microorganisms improved.

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REFERENCES

1. Ledeboer NA. Automation advances microbiology to operational excellence. MLO Med Lab Obs. 2016;48(5):8-11.

2. Froment P, Marchandin H, Vande Perre P, Lamy B. Automated versus manual sample inoculations in routine clinical microbiology: a performance evaluation of the fully automated inoqula instrument. J Clin Microbiol. 2014;52(3):796-802. http://dx.doi.org/10.1128/JCM.02341-13

3. Fontanals D, Sanfeliu L, Pons L, Mariscal D, Torra M. Evaluation of the BacT/Alert and VITAL blood culture systems for the diagnosis of bacteremia. Clin Microbiol Infect. 1998;4(2):88-93. https://dx.doi.org/10.1111/j.1469-0691.1998.tb00361.x

4. Jacobs MR, Mazzulli T, Hazen KC, Good CE, Abdelhamed AM, Lo P, et al. Multicenter clinical evaluation of BacT/Alert Virtuo blood culture system. J Clin Microbiol. 2017;55(8):2413-21. https://dx.doi.org/10.1128/JCM.00307-17

5. Perez KK, Olsen RJ, Musick WL, Cernoch PL, Davis JR, Land GA, et al. Integrating rapid pathogen identification and antimicrobial stewardship significantly decreases hospital costs. Arch Pathol Lab Med. 2013;137(9):1247-54. https://dx.doi.org/10.5858/arpa.2012-0651-OA

6. Pliakos EE, Andreatos N, Shehadeh F, Ziakas PD, Mylonakis E. The costeffectiveness of rapid diagnostic testing for the diagnosis of bloodstream infections with or without Antimicrobial Stewarship. Clin Microbiol Rev. 2018;31(3):e00095-17. https://dx.doi.org/10.1128/CMR.00095-17

7. Dauwalder O, Landrieve L, Laurent F, Montclos M, Vandenesch F, Lina G. Does bacteriology laboratory automation reduce time to results and increase quality management? Clin Microbiol Infect. 2016;22(3):236-43. https://dx.doi.org/10.1016/j.cmi.2015.10.037

8. Croxatto A, Prod'hom G, Faverion F, Rochais Y, Greub G. Laboratory automation in clinical bacteriology: what system to choose? Clin Microbiol Infect. 2016;22(3):217-35. https://dx.doi.org/10.1016/j.cmi.2015.09.030

9. Ling TKW, Liu ZK, Cheng AFB. Evaluation of the VITEK 2 system for rapid direct identification and susceptibility testing of Gram-negative bacilli from positive blood cultures. J Clin Microbiol. 2003;41(10):4705-7. https://dx.doi.org/10.1128/JCM.41.10.4705-4707.2003

10. Nimer NA, Al-Saa'da RJ, Abuelaish O. Accuracy of the VITEK(r)2 system for a rapid and direct identification and susceptibility testing of Gram-negative rods and Gram-positive cocci in blood samples. East Mediterr Health J. 2016;22(3):193-200.

11. Mitchell SL, Alby K. Performance of microbial identification by MALDI-TOF MS and susceptibility testing by VITEK 2 from positive blood cultures after minimal incubation on solid media. Eur J Clin Microbiol Infect Dis. 2017;36(11):2201-6. https://dx.doi.org/10.1007/s10096-017-3046-0

12. Huang AM, Newton D, Kunapuli A, Gandhi TN, Washer LL, Isip J, et al. Impact of rapid organism identification via matrix-assisted laser desorption/ionization time-of-flight combined with antimicrobial stewardship team intervention in adult patients with bacteremia and candidemia. Clin Infect Dis. 2013;57(9):1237-45. https://dx.doi.org/10.1093/cid/cit498

13. Delport JA, Strikwerda A, Armstrong A, Schaus D, John M. MALDI-ToF short incubation identification from blood cultures is associated with reduced length of hospitalization and decrease in bacteremia associated mortality. Eur J Clin Microbiol Infect Dis. 2017;36(7):1181-6. https://dx.doi.org/10.1007/s10096-017-2906-y

14. Cockerill FR, Wilson JW, Vetter EA, Goodman KM, Torgerson CA, Harmsen WS, et al. Optimal testing parameters for blood cultures. Clin Infect Dis. 2004;38(12):1724-30. https://dx.doi.org/10.1086/421087

15. Mutters NT, Hodiamont CJ, Jong MD, Overmeijer HP, van den Boogaard M, Visser CE. Performance of kiestra total laboratory automation combined with MS in clinical microbiology practice. Ann Lab Med. 2014;34(2):111-7. https://dx.doi.org/10.3343/alm.2014.34.2.111

16. Croxatto A, Dijkstra K, Prod'hom G, Greub G. Comparison of inoculation with the InoqulA and WASP automated systems with manual inoculation. J Clin Microbiol. 2015;53(7):2298-307. https://dx.doi.org/10.1128/JCM.03076-14

17. Guo L, Ye L, Zhao Q, Ma Y, Yang J, Luo Y. Comparative study of MALDI-TOF MS and VITEK 2 in bacteria identification. J Thorac Dis. 2014;6(5):534-8. https://dx.doi.org/10.3978/j.issn.2072-1439.2014.02.18

18. Febbraro F, Rodio DM, Puddioni G, Antonelli G, Pietropaolo V, Trancassini M. MALDI-TOF MS Versus VITEK(r) 2: Comparison of systems for the identification of microorganisms responsible for bacteremia. Curr Microbiol. 2016;73(6):843-50. https://dx.doi.org/10.1007/s00284-016-1121-x

19. Doern GV, Vautour RV, Gaudet M, Levy B. Clinical impact of rapid in vitro susceptibility testing and bacterial identification. J Clin Microbiol. 1994;32(7):1757-62.

20. Maurer FP, Christner M, Hentschke M, Rohde H. Advances in rapid identification and susceptibility testing of bacteria in the clinical microbiology laboratory: implications for patient care and antimicrobial stewardship programs. Infect Dis Rep. 2017;9(1):6839. https://dx.doi.org/10.4081/idr.2017.6839

21. Pfaller MA, Boyken L, Hollis RJ, Kroeger J, Messer SA, Tendolkar S, et al. Wild-Type MIC Distributions and epidemiological cutoff values for the echinocandins and Candida spp. J Clin Microbiol. 2010;48(1):52-6. https://dx.doi.org/10.1128/JCM.01590-09

22. Fernandes SC, Dhanashree B. Drug resistance & virulance determinants in clinical isolates of Enterococcus species. Indian J Med Res. 2013;137(5):981-5.

23. Harbarth S, Garbino J, Pugin J, Romand JA, Lew D, Pittet D. Inappropriate initial antimicrobial therapy and its effect on survival in a clinical trial of immunomodulating therapy for severe sepsis. Am J Med. 2003;115(7):529-35. https://dx.doi.org/10.1016/j.amjmed.2003.07.005

24. Micek ST, Lloyd AE, Ritchie DJ, Reichley RM, Fraser VJ, Kollef MH. Pseudomonas aeruginosa bloodstream infection: importance of appropriate initial antimicrobial treatment. Antimicrob Agents Chemother. 2005;49(4):1306-11. https://dx.doi.org/10.1128/AAC.49.4.1306-1311.2005

25. Höffken G, Niederman MS. Nosocomial pneumonia: the importance of a de-escalating strategy for antibiotic treatment of pneumonia in the ICU. Chest. 2002;112(6):2183-96. https://dx.doi.org/10.1378/chest.122.6.2183

26. Soo Hoo GW, Wen YE, Nguyen TV, Goetz MB. Impact of clinical guidelines in the management of severe hospital- acquired pneumonia. Chest. 2005;128(4):2778-87. https://dx.doi.org/10.1378/chest.128.4.2778

27. Rhodes A, Evans LE, Alhazzani W, Levy MM, Antonelli M, Ferrer R, et al. Surviving sepsis campaign: international guidelines for management of sepsis and septic shock: 2016. Intensive Care Med. 2017;43(3):304-77. https://dx.doi.org/10.1007/s00134-017-4683-6