

BRIEF COMMUNICATION

***Pseudomonas* spp. ISOLATED FROM THE ORAL CAVITY OF HEALTHCARE WORKERS FROM AN ONCOLOGY HOSPITAL IN MIDWESTERN BRAZIL**

Ana Beatriz Mori LIMA(1), Lara Stefânia Netto de Oliveira LEÃO-VASCONCELOS(2), Dayane de Melo COSTA(3), Larissa Oliveira Rocha VILEFORT(3), Maria Cláudia Dantas Porfírio Borges ANDRÉ(2), Maria Alves BARBOSA(3) & Marinésia Aparecida PRADO-PALOS(3)

SUMMARY

This cross-sectional study, performed in an oncology hospital in Goiania, aimed to characterize the prevalence of oral colonization and antimicrobial susceptibility of *Pseudomonas* spp. isolated from the saliva of healthcare workers. Microorganisms were subjected to biochemical tests, susceptibility profile, and phenotypic detection. Of 76 participants colonized with Gram negative bacilli, 12 (15.8%) harbored *Pseudomonas* spp. Of all isolates, *P. aeruginosa* (75.0%), *P. stutzeri* (16.7%), and *P. fluorescens* (8.3%), were resistant to cefoxitin, and therefore likely to be AmpC producers. The results are clinically relevant and emphasize the importance of surveillance to minimize bacterial dissemination and multiresistance.

KEYWORDS: *Pseudomonas*; Oral colonization; Healthcare workers.

Pseudomonas spp. is an ubiquitous microorganism frequently isolated in immunocompromised patients. The intrinsic resistance to multiple antimicrobial agents contributes to the opportunistic character of this pathogen. The emergence of healthcare-associated infection (HCAI) is considered a challenge to public health^{1,5,6,8}. The problem increases when AmpC-producing *Pseudomonas* spp. are isolated from asymptomatic professionals. The enzyme confers resistance to cephalosporins and penicillins. The *bla*_{AmpC} genes are found in the chromosome of *Pseudomonas* and CESP group (*Citrobacter*, *Enterobacter*, *Serratia*, and *Providencia*). Bacteria can eventually present sensitivity to cefoxitin, whereas mutant strains express resistance to it due to the loss of repressor genes^{7,9,10,13,14}. Asymptomatic carriers play a fundamental role in the epidemiology of HAI, since they act as disseminators through droplets of saliva expelled during hospital assistance. The asymptomatic colonization may progress to lower respiratory tract infection (pneumonia) and result in health problems to workers and users attended. It is important to establish control strategies to prevent the spread of nosocomial infections. However, studies of the oropharyngeal colonization of healthcare professionals by *Pseudomonas* spp. are scant and the relevance of these carriers should be enlightened^{3,4,11,12}. This research aimed to determine the prevalence of oral colonization and the antimicrobial susceptibility of *Pseudomonas* spp. isolated from the saliva of healthcare workers from an oncology institution located in Goiania, Brazil, reference center in cancer treatment.

This cross-sectional study, carried out from May 2009 to November 2010, involved 294 participants (149 members of the healthcare team and 145 employees from the technical support area) and was approved by the Ethical Committee of the *Associação de Combate ao Câncer de Goiás* (ACCG/040/08). This research is part of a surveillance project Alpha developed in a partnership with some academic units of the *Universidade*

Federal de Goiás (UFG). The study population was composed of workers from the following areas: surgical center, infection control, endoscopy, emergency, chemotherapy, rehabilitation and physiotherapy, radiotherapy, intensive care unit, bone marrow transplantation unit, cleaning, and laundry. Interviews and saliva sampling were performed after participants had signed the informed consent. Unstimulated saliva samples were collected in sterile tubes, transported to the laboratory, and streaked on MacConkey agar. Gram-negative bacilli (GNB) isolates were identified by standardized biochemical tests¹⁵. Susceptibility tests were performed using the diffusion method with Oxoid disks. Disk-approximation method with cefoxitin was used as a phenotypic marker for the detection of probable AmpC producing *Pseudomonas* spp. The results were interpreted according to CLSI guidelines². *Pseudomonas aeruginosa* ATCC 27853 was used as quality control².

Of the 294 participants, 76 were colonized with GNB, and among these, 12 (15.8%) harbored cefoxitin-resistant *Pseudomonas* spp. (100.0%), suggesting AmpC production. Isolates were sensitive to other antimicrobial agents tested (amikacin, aztreonam, cefepime, ceftazidime, ciprofloxacin, imipenem, levofloxacin, meropenem, and piperacillin-tazobactam). Simultaneous colonization by *Pseudomonas* spp. and *Enterobacteriaceae* was observed in two professionals. The prevalence of oral colonization with *Pseudomonas* spp. was higher among cleaning and laundry workers (25.0%), physicians and nurse technicians (16.7%), and radiology technicians (8.3%). Of the workers colonized with cefoxitin-resistant *Pseudomonas* spp. (n = 12), 66.7% were aware of multiresistance, 25.0% used oral antiseptics and 8.3% practiced self-medication. The isolated species were: *P. aeruginosa* (75.0%), *P. stutzeri* (16.7%), and *P. fluorescens* (8.3%). These results have clinical relevance, since these pathogens do not belong to the normal human oral flora and

(1) Secretaria de Estado da Saúde, Seção de Microbiologia, Laboratório Central de Saúde Pública (LACEN), Goiânia, GO, Brazil. E-mail: anabeatrizmori@yahoo.com.br

(2) Universidade Federal de Goiás (UFG), Instituto de Patologia Tropical e Saúde Pública, Departamento de Microbiologia, Goiânia, GO, Brazil. E-mails: larastefania@yahoo.com.br; mcporfírio@hotmail.com

(3) Universidade Federal de Goiás (UFG), Faculdade de Enfermagem, Goiânia, GO, Brazil. E-mails: daynesaga@yahoo.com.br; larissavilefort@gmail.com; maria.malves@gmail.com; marinesiaprado@gmail.com

Correspondence to: Ana Beatriz Mori Lima, Secretaria de Estado da Saúde, Laboratório de Saúde Pública (LACEN), Alameda do Contorno 3556, Jardim Bela Vista, 74853-120 Goiânia, GO, Brazil. Phone: 556232019629. E-mail: anabeatrizmori@yahoo.com.br

may cause health problems, such as tonsillitis, esophagitis, pharyngitis, pneumonia, and other systemic diseases.

It is necessary to point out that although the isolates showed sensitivity to ceftazidime *in vitro*, they can express induced resistance when cephalosporin is administered *in vivo*. Therefore, the use of beta-lactam agents should be discouraged^{7,9,10,13,14}. As previously stated, this study is one of the few aiming to assess the prevalence of GNB and *Pseudomonas* spp. isolated from the oral cavity of health professionals. Investigating 100 healthy people, CONTI *et al.*³ found that 3.2% were colonized with *P. aeruginosa* on the tongue dorsum and, among these, four showed simultaneous colonization by *Pseudomonas* spp. and other GNB, confirming our data. The global prevalence of workers colonized with *Pseudomonas* spp. in the present study was 4.08% (12/294). PRADO-PALOS *et al.*¹¹ evaluated the prevalence of GNB isolated from the saliva of 278 health workers in a teaching hospital in Goiania. A total of 319 GNB were identified, namely 208 (65.2%) *Enterobacteriaceae* and 111 (34.8%) non-fermenters, of which 25 (7.8%) were *P. aeruginosa*, results that are in accordance with our findings. RAMOS *et al.*¹² carried out a study from 1998 to 2008 in Sao Paulo to survey the prevalence of beta-lactam-resistant enteric bacteria and *Pseudomonas* spp. isolated from the oral cavity of 250 patients. The authors isolated 154 GNB, encompassing *P. aeruginosa* (15 isolates) and *P. fluorescens* (four isolates). Susceptibility tests showed that eight (53.3%) isolates of *P. aeruginosa* were resistant to cefoxitin and two (13.3%) to carbapenems. Our results are similar to these findings. MORTARI *et al.*⁹ reported that *P. aeruginosa* was isolated from 90 clinical samples in a research developed in Southern Brazil. Phenotypic detection showed that 31 strains (34.4%) were probable AmpC producers.

Surveillance of the oral colonization of workers by *Pseudomonas* spp. is essential in order to minimize the emergence of HAI. It is important to emphasize that asymptomatic carriers act as disseminators. Strategies are indispensable to determine the prevalence of colonization, monitor the circulating profile, and allow therapeutic interventions. Furthermore, they can ensure excellence in the quality of the service and reduce health hazards to employees, patients, and the community.

RESUMO

***Pseudomonas* spp. isoladas da cavidade oral de profissionais da saúde de um hospital oncológico do centro-oeste do Brasil**

Este estudo transversal, realizado em um hospital oncológico em Goiânia, objetivou caracterizar a prevalência da colonização oral e suscetibilidade antimicrobiana de *Pseudomonas* spp. isoladas da saliva de profissionais da saúde. Micro-organismos foram submetidos a testes bioquímicos, perfil de sensibilidade e detecção fenotípica. Dos 76 participantes colonizados por bastonetes Gram negativos, 12 (15,8%) albergavam *Pseudomonas* spp. Todos os isolados, *P. aeruginosa* (75,0%), *P. stutzeri* (16,7%) e *P. fluorescens* (8,3%), foram resistentes à cefoxitina e, portanto, possíveis produtores de AmpC. Os resultados são clinicamente relevantes e enfatizam a importância da vigilância para minimizar a disseminação e a multirresistência bacteriana.

ACKNOWLEDGEMENTS

The authors thank Fabiana C. Pimenta (Centers for Disease Control and Prevention) for the scientific advice, Nádia F. Gonçalves for the

technical assistance, and the *Fundação de Amparo à Pesquisa do Estado de Goiás* (FAPEG) for the financial support.

CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

REFERENCES

1. Boyer A, Doussau A, Thiébaud R, Venier AG, Tran V, Boulestreau H, *et al.* *Pseudomonas aeruginosa* acquisition on an intensive care unit: relationship between antibiotic selective pressure and patients' environment. *Crit Care*. 2011;15:1-10.
2. Clinical and Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing. 19th Informational Supplement. Wayne: CLSI/document (M100-S19); 2009.
3. Conti S, Santos SSF, Koga-Ito CY, Jorge AOC. *Enterobacteriaceae* and *Pseudomonadaceae* on the dorsum of human tongue. *J Appl Oral Sci*. 2009;17:375-80.
4. Fortaleza CMCB, Figueiredo LC, Beraldo CC, Melo EC, Póla PMS, Aragão VDN. Risk factors of oropharyngeal carriage of *Pseudomonas aeruginosa* among patients from a medical-surgical intensive care unit. *Braz J Infect Dis*. 2009;13:173-6.
5. Gonçalves DCPS, Lima ABM, Leão LSNO, Carmo Filho JR, Pimenta FC, Vieira JDG. Detecção de metalo-beta-lactamase em *Pseudomonas aeruginosa* isoladas de pacientes hospitalizados em Goiânia, Estado de Goiás. *Rev Soc Bras Med Trop*. 2009;42:411-4.
6. Gudiol C, Tubau F, Calatayud L, Garcia-Vidal C, Ciscal M, Sánchez-Ortega I, *et al.* Bacteremia due to multidrug-resistant Gram-negative bacilli in cancer patients: risk factors, antibiotic therapy and outcomes. *J Antimicrob Chemother*. 2011;66:657-63.
7. Jacoby GA. AmpC beta-lactamases. *Clin Microbiol Rev*. 2009;22:161-82.
8. Kobayashi CCBA, Sadoyama G, Vieira, JDG. Determinação da resistência antimicrobiana associada em isolados clínicos de *Staphylococcus aureus* e *Pseudomonas aeruginosa* em um hospital público de Goiânia, Estado de Goiás. *Rev Soc Bras Med Trop*. 2009;42:404-10.
9. Mortari AP, Saucedo EM, Pereira RS, Duarte M, Vizzotto BS, Santos RCV. Prevalência de *Pseudomonas aeruginosa* produtoras de β -lactamases do tipo AmpC em isolados clínicos de Santa Maria, RS. *Rev Bras Anal Clin*. 2008;40:147-9.
10. Pfeifer Y, Cullik A, Witte W. Resistance to cephalosporins and carbapenems in Gram-negative bacterial pathogens. *Int J Med Microbiol*. 2010;300:371-9.
11. Prado-Palos MA, Gir E, Lima ABM, Leão LSNO, Pimenta FC. Prevalência de bastonetes Gram-negativos isolados da saliva de trabalhadores da saúde. *Rev Eletr Enferm*. 2011;13:730-4.
12. Ramos MMB, Gaetti-Jardim EC, Gaetti-Jardim Junior E. Resistance to tetracycline and β -lactams and distribution of resistance markers in enteric microorganisms and pseudomonads isolated from the oral cavity. *J Appl Oral Sci*. 2009;17:13-8.
13. Rodríguez-Martínez JM, Poirel L, Nordmann P. Extended-spectrum cephalosporinases in *Pseudomonas aeruginosa*. *Antimicrob Agents Chemother*. 2009;53:1766-71.
14. Upadhyay S, Sen MR, Bhattacharjee A. Presence of different beta-lactamase classes among isolates of *Pseudomonas aeruginosa* expressing AmpC beta-lactamase enzyme. *J Infect Dev Ctries*. 2010;4:239-42.
15. Winn WC Jr, Allen SD, Janda WM, Koneman EW, Procop GW, Schreckenberger PC, *et al.* Koneman's color atlas and textbook of diagnostic microbiology. 6th ed. Philadelphia: Lippincott Williams and Wilkins; 2008.

Received: 12 October 2014
Accepted: 16 February 2015