



Letter to the Editor

Can the resistance profile affect the survival of *Acinetobacter baumannii* on hospital surfaces?

Sir,

The role of the inanimate environment as a reservoir or source of transmission of nosocomial pathogens is well recognized [1–3]. Although the survival of *Acinetobacter baumannii* on hospital surfaces is an important factor in the transmission chain, the link between resistance to antibiotics and resistance to desiccation in *A. baumannii* has received little research attention [4,5].

We assessed the survival of *A. baumannii* of different clonal complexes (CC) and resistance profiles (resistant, multi-drug-resistant and extensively drug-resistant) on eight dry surfaces that are representative of the hospital environment for 300 days.

Three different clinical isolates of *A. baumannii*, typed by multi-locus sequence typing, were selected: AcR/CC79 (*A. baumannii* resistant ST79, CC79; carbapenem sensitive); AcMDR/CC15 (*A. baumannii* multi-drug-resistant ST15, CC15; carbapenem resistant and polymyxin sensitive); and AcXDR/CC1 (*A. baumannii* extensively drug-resistant ST983, CC1; carbapenem/polymyxin resistant). The following test surfaces were used: polyurethane (Mattress A), nappa (Mattress B), stainless steel, paper, vinyl flooring, computer keys, fabric (cotton) and glass.

Initially, test surfaces (TSs) measuring 10 × 20 mm were washed, placed in flasks with distilled water (Mattresses A and B, stainless steel, vinyl flooring, glass and computer keys) or without water (paper and fabric), sterilized in an autoclave for 15 min at 121°C, and allowed to dry in sterile Petri dishes in a biological safety cabinet.

The isolates were grown for 18–24 h on nutrient agar in Petri dishes, and colonies of each were inoculated in saline (0.85% NaCl). To standardize the density of the inoculum used to artificially contaminate the TSs, the saline prepared with the bacterial suspension was compared with a BaSO₄ turbidity standard, equivalent to the 0.5 MacFarland standard [bacterial suspension containing approximately 1–2 × 10⁸ colony-forming units (cfu)/mL]. Ten-microlitre volumes of these suspensions were applied to the TSs (approximately 10⁶ cfu/TS) in triplicate. After the inoculum dried, the TSs were transferred to 150 × 15-mm sterile Petri dishes and kept in the laboratory for

300 days. The mean temperature was 26.3°C and the mean relative humidity was 66.6% during the experiments.

The surviving bacteria were recovered by transferring the contaminated TS to tubes that contained 5 mL of brain-heart infusion broth (10 mL for the computer keys) immediately after the inoculum dried (time zero) and daily for 60 days, and then every five days until the end of the experiment. The total number of contaminated test surfaces was 7776 (2592 surfaces per *A. baumannii* strain). Student's *t*-test was used to compare the survival of the different isolates of *A. baumannii* on the test surfaces.

All tested isolates of *A. baumannii* survived for at least 14 days on the test surfaces (Figure 1). Except on glass, vinyl flooring and polyurethane (Mattress A), AcXDR/CC1 (carbapenem/polymyxin resistant) showed the highest resistance to desiccation, remaining viable for 113 days on nappa (Mattress B), 92 days on paper, 75 days on cotton fabric, 51 days on computer keys, and 41 days on stainless steel. In contrast, AcMDR/CC15 (carbapenem resistant and polymyxin sensitive) and AcR/CC79 (carbapenem sensitive) survived for longer on vinyl floor (36 days) and polyurethane (Mattress A, 41 days), respectively (Figure 1). There were significant differences between the survival of AcXDR/CC1 and AcMDR/CC15.

Interestingly, the polymyxin-resistant *A. baumannii* survived longer on different surfaces compared with the isolates sensitive to polymyxin. This is worrying because AcXDR/CC1 belongs to CC1, which is often associated with epidemic spread, and outbreak strains are frequently multi-drug resistant [6].

The multiple mechanisms of antibiotic resistance found in *A. baumannii* may play a role in its survival in the environment [7]. In the case of polymyxin resistance, for example, observations by Boll *et al.* [8] suggested that hepta-acylation of lipid A fortifies the outer membrane to protect *A. baumannii* from cationic antimicrobial peptides and desiccation. This may explain the higher survival of the polymyxin-resistant *A. baumannii* strain (AcXDR/CC1) in this study.

A limitation of this study is the small sample size. However, to our knowledge, this is the first demonstration that polymyxin resistance is associated with longer survival periods of *A. baumannii* on surfaces typical of the hospital environment. Further studies testing a larger number of *A. baumannii* strains are required to confirm these findings. Another factor to be considered is that different ambient temperature and humidity conditions may cause significant variations in the survival profile of *A. baumannii* on dry surfaces [4]. As the present study was conducted at high humidity (66.6%) and temperature (26.3°C), the findings may be readily applicable in countries with similar climates, but not in colder and/or drier regions.

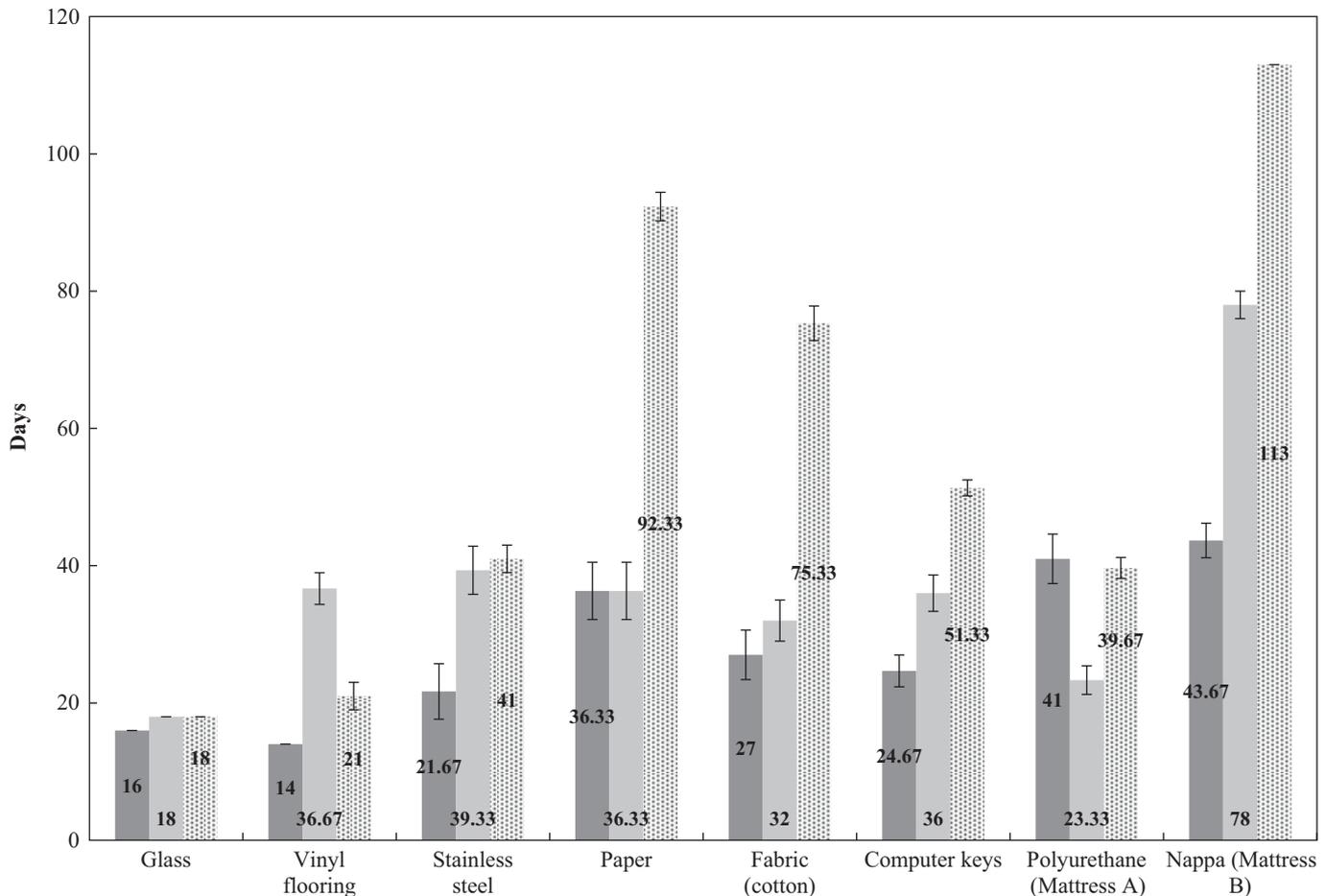


Figure 1. Survival of the three *Acinetobacter baumannii* isolates on eight different materials. Dark grey bars, AcR/CC79; light grey bars, AcMDR/CC15; stippled bars, AcXDR/CC1.

In conclusion, the strains of *A. baumannii* investigated survived for long periods on inanimate surfaces, suggesting that the type of material and antibiotic resistance may influence the survival of *A. baumannii* in the environment.

Conflict of interest statement

None declared.

Funding sources

None.

References

- [1] Kramer A, Schwebke I, Kampf G. How long do nosocomial pathogens persist on inanimate surfaces? A systematic review. *BMC Infect Dis* 2006;6:130.
- [2] Otter JA, Yezli S, French GL. The role played by contaminated surfaces in the transmission of nosocomial pathogens. *Infect Control Hosp Epidemiol* 2011;32:687–99.
- [3] Cheng VCC, Wong S-C, Chen JHK, So SYC, Wong SCY, Ho P-L, et al. Control of multidrug-resistant *Acinetobacter baumannii* in Hong Kong: role of environmental surveillance in communal areas after a hospital outbreak. *Am J Infect Control* 2018;46:60–6.
- [4] Jawad A, Heritage J, Snelling AM, Gascoyne-Binzi DM, Hawkey PM. Influence of relative humidity and suspending menstrua on survival of *Acinetobacter* spp. on dry surfaces. *J Clin Microbiol* 1996; 34:2881–7.
- [5] Weber DJ, Rutala WA. Understanding and preventing transmission of healthcare-associated pathogens due to the contaminated hospital environment. *Infect Control Hosp Epidemiol* 2013; 34:449–52.
- [6] Diancourt L, Passet V, Nemeč A, Dijkshoorn L, Brisse S. The population structure of *Acinetobacter baumannii*: expanding multi-resistant clones from an ancestral susceptible genetic pool. *PLoS One* 2010;5:e10034.
- [7] Greene C, Vadlamudi G, Newton D, Foxman B, Xi C. The influence of biofilm formation and multidrug resistance on environmental survival of clinical and environmental isolates of *Acinetobacter baumannii*. *Am J Infect Control* 2016;44:e65–71.
- [8] Boll JM, Tucker AT, Klein DR, Beltran AM, Brodbelt JS, Davies BW, et al. Reinforcing lipid A acylation on the cell surface of *Acinetobacter baumannii* promotes cationic antimicrobial peptide resistance and desiccation survival. *mBio* 2015;6:e00478–15.

F.G. Lodi
G.F. Viana
A.P. Uber
N.H. Fedrigo
A.P. Montemezzo de Farias
M.M. dos Anjos Szczerepa
C.L. Cardoso

S.A.B. Nishiyama
M.C.B. Tognim*

*Laboratório de Microbiologia, Departamento de Ciências
Básicas da Saúde, Universidade Estadual de Maringá, Maringá,
Paraná, Brazil*

Universidade Estadual de Maringá, Avenida Colombo 5790. CEP
87020-900, Maringá, Paraná, Brazil. Tel.: +55 44 3011 4952;

fax: +55 44 3011 5941.

E-mail address: mcbtognim@gmail.com.br (M.C.B. Tognim)

Available online 16 March 2019

* Corresponding author. Address: Laboratório de
Microbiologia, Departamento de Ciências Básicas da Saúde,