

ACINETOBACTER BAUMANNII FROM SEWAGE OF NURSING HOME IN CROATIA

Branka Bedenić^{1,2}, Nataša Beader^{1,2}, Marko Siroglavić³, Mia Slade⁴, Haris Car⁵, Svjetlana Dekić⁶,
Martina Šeruga Musić⁶, Aleksandra Presečki-Stanko², Dorotea Šijak¹, Jasna Hrenović⁶

¹Department of Microbiology, School of Medicine, University of Zagreb, Zagreb, Croatia; ²Clinical Department of Clinical and Molecular Microbiology, University Hospital Centre Zagreb, Zagreb, Croatia; ³Clinical Department of Transfusion Medicine and Transplantation Biology, University Hospital Centre Zagreb, Zagreb, Croatia; ⁴Croatian Institute for Transfusion Medicine, Zagreb, Croatia; ⁵Secondary Medical School, Zagreb, Croatia; ⁶Faculty of Science, University of Zagreb, Zagreb, Croatia

Correspondence: Branka Bedenić, Clinical Department for Clinical and Molecular Microbiology, School of Medicine, University of Zagreb, University Hospital Center Zagreb, tel: +385 1 23 67 304, fax: +385 1 4590 130, e-mail: branka.bedenic@kbc-zagreb.hr; bbedenic@mef.hr

Background

In the previous studies the predominance of OXA-24/40 and OXA-23 β -lactamase was reported among *Acinetobacter baumannii* in both the hospitals and long-term care facilities (LTCF) in Croatia. The aim of this study was to determine antibiotic susceptibility, mechanisms of antibiotic resistance and molecular epidemiology of *A. baumannii* isolates recovered from sewage of a nursing home in Zagreb.

Materials/methods

- In April 2017, 10 *A. baumannii* were isolated from sewage.
- Antibiotics susceptibility was determined by broth microdilution method.
- The presence of carbapenemases of class A, B and D and extended-spectrum β -lactamases genes was explored by PCR.
- The occurrence of the ISAbal1 upstream of the *bla*_{OXA-51-like} or *bla*_{OXA-23-like} was determined by PCR mapping.
- Conjugation and transformation experiments were performed as previously described. Plasmid incompatibility groups were determined by PBRT.
- Genotyping were performed by sequence group determination, PFGE and MLST.

Results

Eight isolates were resistant to gentamicin and ciprofloxacin, three to ceftazidime and cefepime and two to carbapenems (Table 1). There was uniform susceptibility to colistin (Table 1). All isolates were positive for chromosomal *bla*_{OXA-51} whereas two carbapenem-resistant isolates were found to possess also *bla*_{OXA-23} genes. ISAbal1 was found upstream of *bla*_{OXA-51} in seven out of ten isolates. Attempts to transfer imipenem resistance were unsuccessful indicating chromosomal location of *bla*_{OXA-23} gene. The plasmids extracted from isolates positive for OXA-23 belonged to Inc group 6 encoding *aci6*-replicase gene.

Two OXA-23 positive isolates belonged to SG 1 (IC2) whereas the rest of the isolates susceptible to carbapenems were allocated to SG 2 (IC1). Additional MLST analysis corroborated that two OXA-23 and carbapenem-resistant isolates belong to the ST-195 clustering into the CC92 within the IC2, commonly reported worldwide. The susceptible isolates belonging to IC1 were found to be clonally related by PFGE, but different from the carbapenem-resistant (Fig 1).

Table 1.

Antibiotic susceptibility, β -lactamase production and genotypes of *A. baumannii* isolates from nursing home sewage water

Strain number	Date of isolation	Minimum inhibitory concentrations (MIC) mg/L												IC	PFGE and ST	
		CAZ	CTX	CRO	FEP	TZP	SAM	IMI	MEM	GM	CIP	TGC	COL			BL
Poli 1 (P1)	24.4.2017	1	>128	>128	1	16	8	0.12	0.25	1	0.25	1	0.5	OXA-51-like	IC I	la
Godan 1 (G1)	24.4.2017	2	>128	>128	1	8	4	0.25	0.25	0.5	0.12	1	1	OXA-51-like	IC I	lb
Godan 2 (G2)	24.4.2017	>128	>128	>128	>128	>128	16	0.5	2	>128	>128	2	1	OXA-51-like	IC I	S
Kvatrić 1 (K1)	2.5.2017	>128	>128	>128	>128	>128	16	32	>128	>128	>128	0.5	2	OXA-66, OXA-23	IC II	S, ST195
Kvatrić 2 (K2)	2.5.2017	16	>128	>128	16	64	4	0.5	1	>128	>128	0.5	1	OXA-51-like	IC I	lc
Kvatrić 3 (K3)	2.5.2017	32	>128	>128	8	64	4	0.5	1	>128	>128	0.5	1	OXA-51-like	IC I	le
Kvatrić 4 (K4)	2.5.2017	>128	>128	>128	>128	>128	8	32	>128	>128	>128	0.5	2	OXA-66, OXA-23	IC II	S, ST195
Kvatrić 5 (K5)	2.5.2017	2	>128	>128	1	16	2	0.5	2	>128	>128	2	0.5	OXA-51-like	IC I	ld
Kvatrić 6 (K6)	2.5.2017	4	>128	>128	2	16	8	0.5	2	>128	>128	1	0.5	OXA-51-like	IC I	lb
Kvatrić 7 (K7)	2.5.2017	4	>128	>128	1	32	8	0.25	1	>128	>128	2	0.5	OXA-51-like	IC I	lb

Abbreviations: TZP-piperacillin/tazobactam; SAM; sulbactam/ampicillin; CAZ-ceftazidime; CRO-ceftriaxone; FEP-cefepime; IMI-imipenem; MEM-meropenem; GM-gentamicin; CIP-ciprofloxacin; TGC-tigecycline; COL-colistin; ST-sequence type; IC-International clonal lineage; BL: beta-lactamase content

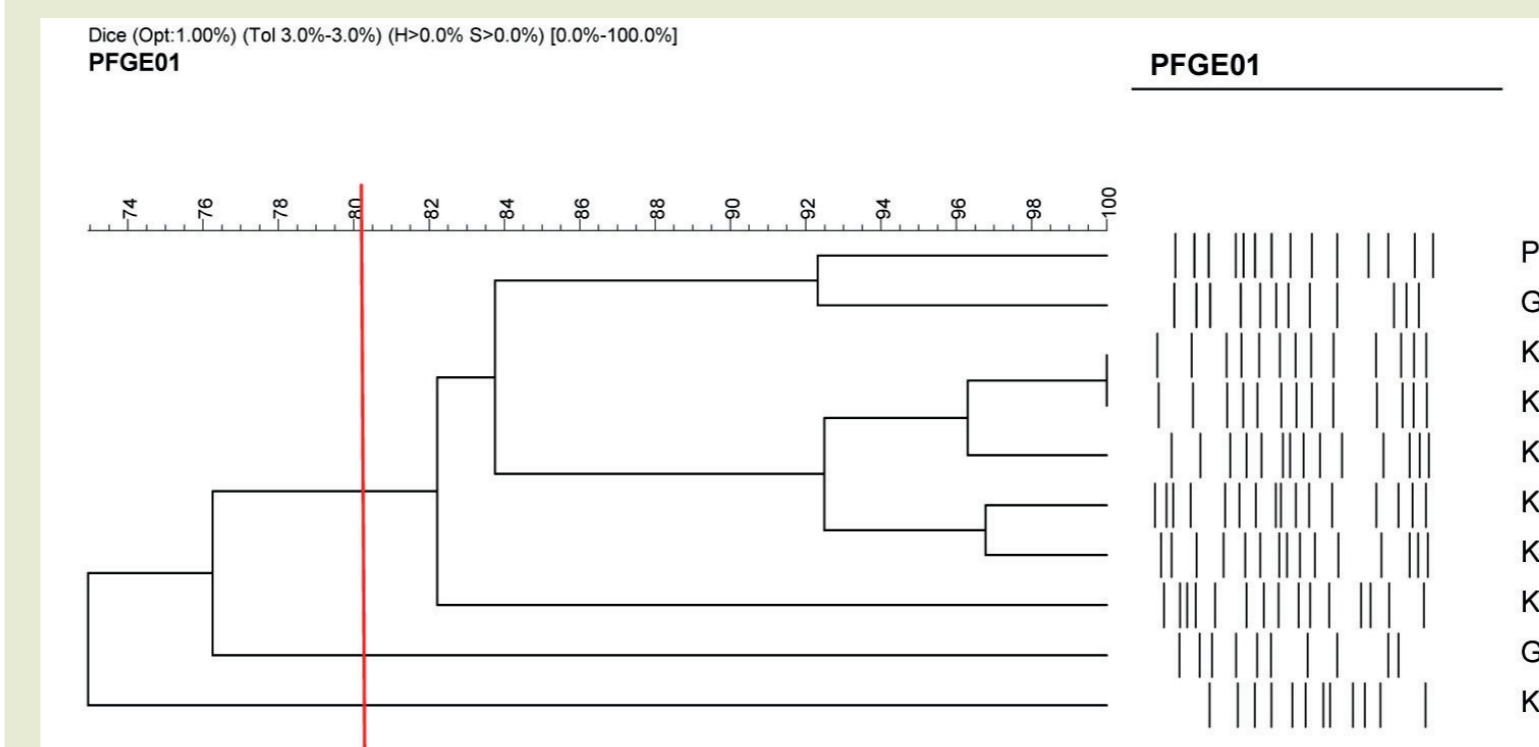


Figure 1. PFGE profiles of *A. baumannii* isolates from nursing home sewage water.

Conclusions

- This study showed dissemination of OXA-23 producing *A. baumannii* from the nursing home into the sewage.
- Disinfection of nursing home sewage should be recommended in order to prevent the spread of resistance genes into the community sewage and nature.
- In 2013, OXA-23 positive *A. baumannii* from investigated nursing home were found to belong to ST-487, demonstrating the emergence of a new clone.