

Molecular Epidemiology of *Providencia* spp. Harboring NDM Carbapenemase in Argentina.

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Background: the incidence of NDM carbapenemase-producing organisms has been increasing in Argentina since 2013, mobilized by the escalation of *Providencia* spp. From 2013-2015 the NRL has confirmed 39 *Providencia* spp. isolates (16 *P. rettgeri* [PRE] and 23 *P. stuartii* [PST]) harboring *bla*_{NDM1}, representing 45% of 86 gram-negative bacilli NDM confirmed cases. Here we describe the molecular epidemiology of these isolates. **Materials/methods:** we designed an algorithm to detect carbapenemases at the level of the clinical microbiology laboratory, which is currently implemented by 432 laboratories across Argentina (National QC Program in Bacteriology, Ministry of Health). By means of this algorithm, isolates with decreased susceptibility to carbapenems and a positive synergy test result between carbapenems and EDTA disks, were considered as suspicious of NDM production and referred to the NRL. Strains were identified using MALDI-TOF. *bla*_{NDM-1} was confirmed by PCR and DNA sequencing and the genetic relatedness between isolates was assessed by PFGE (DNA digested with NotI). Antimicrobial susceptibility was evaluated by disk diffusion and dilution methods (CLSI). Statistic analyses were performed with Statistics Pro. **Results. Table.** PRE was recovered mainly from urine (14/16, 88%) unlike PST (8/23, 35%) ($p < 0.01$). Isolates from blood, skin/soft tissues or respiratory tract corresponded only to PST ($p < 0.01$). Carbapenem non-susceptibility: 100% imipenem and meropenem and 77% ertapenem (100% PRE vs. 61% PST [$p < 0.01$]). PRE and PST isolates segregated in different hospitals, with the exception of one Institution with one strain of each species. 4/14 (29%) hospitals reported PRE or PST during more than one calendar year. 10/39 (26%) isolates co-produced ESBLs (7 CTXM, 3 PER-2). Susceptibility to only one antimicrobial agent (amikacin) was mostly linked to PST (31% vs 6% [$p < 0.01$]). By PFGE, 4 PRE and 5 PST pulse-types were observed. Each dominant clone included 8/16 (3 Hospitals) and 11/23 (5 Hospitals) isolates of PRE and PST, respectively. One hospital concentrated 56% of PRE isolates (pulse-type A and B), unlike those with PST (range: 4-17% per hospital). PST strains belonging to pulse-type A1 (5 Hospitals/4 cities) were associated with increased likelihood of severe infection (bloodstream [$p < 0.01$]), to co-produce CTXM ($p < 0.01$), or to display an extreme-resistant phenotype (only susceptible to amikacin [$p < 0.01$]).

	Year			Total (2013-2015)
	2013	2014	2015	
No. of <i>P. rettgeri</i> <i>bla</i>_{NDM-1} isolates	3	8	5	16
No. of Hospitals	2	1	5	6
No. of Cities	1	1	1	1
No. and type (%) of ESBL coproduction	2 (PER-2)	1 (PER-2)	0	3 (18%)
No. (%) of isolates susceptible to:				
>= 3 drugs	2	6	3	11 (69%)
2 drugs	1	1	2	4 (25%)
1 drug	0	1	0	1 (6%)
PFGE types/sub-types (No. of isolates):	A1 (1), A2 (1), A3 (1)	A1 (1), A4 (1), A5 (1)		A (6) (38%)
		B1 (2), B2 (1), B3 (1), B4 (1)	B2 (1), B3 (1), B5 (1)	B (8) (50%)
			C (1)	C (1) (6%)
			D (1)	D (1) (6%)
No. of <i>P. stuartii</i> <i>bla</i>_{NDM-1} isolates		6	17	23
No. of Hospitals		4	7	9
No. of Cities		4	6	6
No. and type (%) of ESBL coproduction		0	7 (CTXM)	7 (30%)
No. (%) of isolates susceptible to:				
>= 3 drugs		6	6	12 (52%)
2 drugs		0	4	4 (17%)
1 drug		0	7	7 (31%)
PFGE types/sub-types (No. of isolates):			A1 (3), A2 (1), A3 (1), A4 (1)	A (11) (48%)
		B1 (4), B2 (1)	B3 (1), B4 (1)	B (7) (30%)
		C1 (1)	C2 (1), C3 (1)	C (3) (13%)
			D (1)	D (1) (4%)
			E (1)	E (1) (4%)

Conclusions: we confirmed the active circulation of NDM-producing *Providencia* spp. in Argentina. Most PRE isolates resulted from the expansion of two clones within a single hospital. By contrast, PST showed a more complex epidemiology, with the dissemination of two clones in multiple hospitals, 5 and 3 respectively. The escalation during 2015 of a single pulse-type (PST A1) allowing few available therapeutic options is of high concern.