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PERSPECTIVE

Metallo-β-Lactamase (MBL)-Producing *Enterobacteriaceae* in United States Children

Latania K. Logan[1,2] and Robert A. Bonomo[2,3,4]

[1]Pediatrics, Rush University Medical Center, Rush Medical College, Chicago, Illinois; [2]Research Service, Louis Stokes Cleveland Department of Veterans Affairs Medical Center, Ohio; [3]Medicine and [4]Pharmacology, Molecular Biology, and Microbiology, Case Western Reserve School of Medicine, Cleveland, Ohio

Metallo-β-lactamases (MBLs) are emerging as the most notable resistance determinants in *Enterobacteriaceae*. In many cases, the genes encoding MBLs are part of complex, mobile genetic elements that carry other resistance determinants. In the United States, there are increasing reports of MBL-producing *Enterobacteriaceae*, with New Delhi MBLs (NDMs) accounting for the majority of transmissible MBL infections. Many infections caused by NDM-producing bacteria are associated with international travel and medical tourism. However, little recognition of the introduction of MBL-producing *Enterobacteriaceae* into the pediatric community has followed. Reports suggest that this occurred as early as 2002. Here, we reflect on the unwelcome emergence of MBL-producing *Enterobacteriaceae* in US children and the available clinical and molecular data associated with spread. Since 2002, there have been disturbing reports that include the most readily transmissible MBLs, bla_{IMP}, bla_{VIM}, and bla_{NDM} types. In the majority of children with available data, a history of foreign travel is absent.

Keywords. carbapenemase; child; drug resistance; *Enterobacteriaceae*; epidemiology.

Metallo-β-lactamases (MBLs) were first detected in the 1960s; however, it was not until the 1990s when increasing reports of clinical infections and nosocomial outbreaks associated with transmissible MBL genes in Gram-negative bacteria (GNB) sparked global attention. Metallo-β-lactamases are Ambler Class B enzymes requiring zinc for activity. As such, they are able to hydrolyze most β-lactams, including carbapenems, while sparing aztreonam. The *bla* genes encoding MBLs are often embedded in complex integrons, which are associated with transposons or plasmids containing multiple drug resistance genes that can be readily transferred between organisms [1, 2]. Notable acquired MBL genes in GNB include $bla_{IMP\text{-}type}$, $bla_{VIM\text{-}type}$, and $bla_{NDM\text{-}type}$ MBLs. Although there are 3 MBL subclasses (B1–B3) that differ by amino acid sequence homology, almost all of clinically important, acquired MBLs belong to subclass B1 [1, 2].

The first reports of transmissible MBL-producing *Enterobacteriaceae* in the United States were in adults in 2009 ($bla_{VIM\text{-}type}$ and $bla_{NDM\text{-}type}$), although MBL-producing *Pseudomonas aeruginosa* were reported in the United States as early as 2004 (VIM-7) and associated with nosocomial outbreaks in adults in 2005 (VIM-2) [2]. Since that time, there has been a worrisome increase in MBL-producing *Enterobacteriaceae* reported in the United States associated with several infection types and outbreaks. As of April 2016, the Centers for Disease Control and Prevention reported 168 isolates of NDM-type and VIM-type MBL-producing *Enterobacteriaceae* in 27 states, and the majority (151) were NDM-type (http://www.cdc.gov/hai/organisms/cre/TrackingCRE.html#CREmapNDM).

What has gone relatively unrecognized is the dissemination of these organisms in our most vulnerable population, children. Since the discovery of NDM-type MBLs in *Enterobacteriaceae* in 2008 from a patient returning to Sweden after healthcare in India, there have been widespread reports of these organisms in multiple countries, and NDM-type MBLs have become the most prevalent MBL gene circulating worldwide [1]. In India where MBL-producing GNB are endemic, reports of newborn and infant-related deaths caused by multidrug-resistant infections, including NDM-producing *Enterobacteriaceae*, continue to increase. Although a majority of reports suggest that the global spread of bla_{NDM} in GNB is predominantly related to international travel and medical tourism, the horizontal gene transfer of bla_{NDM} in endemic areas occurs outside of hospitals and has been linked to household transmission and exposure from environmental sources, which may play an important role in childhood acquisition of drug-resistant organisms [1].

In the United States, a recent national study of trends of carbapenem-resistant *Enterobacteriaceae* (CRE) in children using antimicrobial susceptibility data from 300 US laboratories reported (1) that 266 isolates were identified as CRE

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Correspondence: L. K. Logan, MD, 1653 W. Congress Parkway, Suite 951 Jelke, Chicago, IL 60612 (latania_logan@rush.edu).

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and (2) that the prevalence of CRE isolated from children increased between the years of 1999 and 2012; however, little is known about the molecular mechanisms associated with these increases [3].

In this commentary, we reviewed the literature of MBL-producing *Enterobacteriaceae* in US children and found extremely disturbing reports [4–9]. In addition, there are 2 additional recent cases for which publications are submitted or in press. We also describe an additional unreported pediatric case in the Study for Monitoring Antimicrobial Resistance Trends (SMART) data (S. Bouchillon, MD, personal communication was written on 12 January 2016). In total, we found that 12 pediatric patients, ages 0–11 years, with 15 MBL-producing *Enterobacteriaceae* isolates recovered during clinical care.

Metallo-β-lactamase-producing *Enterobacteriaceae* were first described in US children in 2011; however, infections with $bla_{\text{IMP-4}}$-producing *Escherichia coli* were discovered in pediatric patients as early as 2002, and an *Enterobacter cloacae* harboring $bla_{\text{VIM-2}}$ was recovered from a child in 2005 (Table 1). Of the 15 MBL-producing isolates, the predominant organism was *Klebsiella pneumoniae* in 9 cases (60%), followed by *E coli* in 5 (33.3%) isolates, and in 1 (6.7%) patient, there was an *E cloacae* harboring an MBL gene. All MBL genes (bla_{IMP}, bla_{VIM}, bla_{NDM}) were associated with more than one species, and of importance, the presence of additional plasmid-borne beta-lactamase genes, including extended-spectrum beta-lactamases ($bla_{\text{CTX-M}}$, bla_{TEM}, bla_{SHV}) and AmpC cephalosporinases (bla_{CMY}), were reported in 50% of MBL harboring patients. Multilocus sequence types (STs) were reported for 6 patients and were associated with different STs. For *K pneumoniae*, a $bla_{\text{VIM-4}}$ was ST14, a bla_{NDM} was ST37, whereas a $bla_{\text{IMP-13}}$ was found in ST253. For *E coli*, the 2 $bla_{\text{IMP-4}}$ were ST648 and ST1061, and the $bla_{\text{NDM-1}}$ was ST101.

The median age of children with MBL-producing *Enterobacteriaceae* was 2.5 years (range, 4 months–11 years); 67% were male, and 83% were cared for in medical centers on the Western Coast of the United States (California and Washington) at the time of infection. Unit-level data were available for 10 patients, and 6 (60%) patients were located in the intensive care unit at the time of the culture. The most common source was urine (40%), followed by blood (26.7%) and respiratory cultures (20%). All positive blood cultures were recovered from central venous catheters.

Clinical data were available for 10 of 12 (83.3%) children. All 10 (100%) suffered from serious underlying medical conditions with 40% having hematologic-oncologic comorbidities or were immunosuppressed (including organ transplant), 40% had a neurologic comorbidity, 30% had respiratory comorbidities (including tracheostomy and ventilator dependence), and 20% had a history of early preterm birth of less than 30 weeks gestation. In each case, there was a previous history of hospitalization and/or frequent medical care, and most reported previous exposure to antibiotics. It is interesting to note that travel outside the United States in 6 (50%) patients, which included 5 cases involving bla_{IMP}, and in 1 of the $bla_{\text{NDM-1}}$ cases was not present. Data on hospital type were available for 7 children, and 6 (85.7%) children were cared for in free-standing children's hospitals.

Treatment and outcome data were available for 11 of 12 children. Antibiotic choices were varied and included combination therapy in 5 (45.4%) children. A carbapenem was used in the treatment of 5 children (45.4%); an aminoglycoside (gentamicin or amikacin) was used in the treatment of 4 (36.3%) children; colistin and trimethoprim-sulfamethoxazole were each used in 2 (18.1%) cases; and fosfomycin, levofloxacin, or tigecycline were part of therapy each in 1 (9%) case. Two deaths were reported, with an attributable mortality of 18% due to infections by MBL-producing organisms.

Metallo-β-lactamase-producing *Enterobacteriaceae* infections have been occurring in US children, but for the most part infections have been "under the radar". Consistent with adult data is the heterogeneity of clonal strains associated with MBL dissemination. As a result, spread is likely attributed to "promiscuous plasmids" of multiple types associated with integrons (and transposons in some cases). What is most shocking is that there are reports of cases of MBL-producing *Enterobacteriaceae* infections in young children as early as 2002, even though the first reports were not published in the United States until 2010. Even more surprising is that half of these organisms were found in children who do not have a history of travel. So where are the children acquiring these infections? Other than the 2 cases of $bla_{\text{IMP-4}}$ with evidence of a neonatal intensive care unit stay overlapping the sentinel case [5], the available evidence does not shed clear light on this. We additionally hypothesize that because there appears to be a higher proportion of isolates in children recovered from the Western region of the United States, that there may have been an introduction of MBLs via Southeast Asia and Japan.

Most disturbingly, the limited literature available suggests that children may be silently harboring MDR *Enterobacteriaceae*, including MBL-producers, and that colonization is often prolonged, which means that children may serve as potential reservoirs for these organisms in the community [10–12]. Why children are more likely to become colonized but not infected is poorly understood. The long-term consequences of this prolonged colonization are unthinkable.

In summary, MBL-producing *Enterobacteriaceae* are a real and present danger in children. Regional and national surveillance, as well as the analysis of molecular mechanisms in MDR *Enterobacteriaceae* are critical to identifying and halting the spread of these organisms in our most vulnerable populations. We posit that a targeted surveillance program should be undertaken in critically ill and immunocompromised children who are cared for in tertiary care settings.

Table 1. Characteristics of 12 Pediatric Patients With 15 MBL-Producing *Enterobacteriaceae* Isolates

Year Isolated[a]	Age, Years	Sex	Race/ Ethnicity	Region	Organism	Source	Unit	MBL Gene	Other *bla* Genes	Underlying Conditions	Travel Outside of United States	Treatment	Outcome	Ref.
2002	3.5	M	White	Seattle, WA	*E coli*	Blood	Heme-onc	IMP-4	CMY-2	AML	None	GNT	Survived	[6]
2003	11	M	Hispanic	Seattle, WA	*E coli*	Blood; Stool	Heme-Onc	IMP-4	CMY-2	ALL	Mexico	GNT, TMP/SMX	Survived	[6]
2005	3	F	ND	Los Angeles, CA	*E cloacae*	Respiratory	PICU	VIM-2	TEM-15, ACT	ND	ND	ND	ND	PC
2009	0.33	M	ND	CA	*K pneumo*	Urine	NICU	IMP-4	NR	Premature birth, (25 wks)	None	NT	Survived	[5]
2010	0.42	F	ND	CA	*K pneumo*	Urine	NICU	IMP-4	NR	Premature birth (29 wks); CHD	None	TMP/SMX	Survived	[5]
2010	0.42	M	ND	CA	*K pneumo*	Urine	ED[b]	IMP-4	NR	NR	None	NT	Survived	[5]
2011	1.1	M	Asian	Los Angeles, CA	*K pneumo*	Respiratory	ED	NDM-1	NR	RAD; CSA; DD, Multi-abx courses	Pakistan	COL	Survived	[7]
2012	4	M	White	Chicago, IL	*K pneumo*	Peritoneal	PICU	IMP-13	None	SBT	None	MEM	Survived	[c]
2012	7	F	Asian	Stanford, CA	*E coli*; *K pneumo*	Urine	NR	NDM-1	NR	Neurogenic bladder	India	MEM, TIG, FOS	Survived	[4]
2012	2	F	Asian	Los Angeles, CA	*E coli*	Blood	NR	NDM-1	CTX-M-15; CMY-42	MDS	India	IMI, AMK	Died	[8]
2013	3	M	NR	Los Angeles, CA	*K pneumo*	Urine; Blood	PICU	NDM-1	CTX-M-15; CMY-4; SHV-11	Gangliosidosis; Trach/Vent dep	None	LEV, IMI, COL	Survived	[8]
2014	2	M	Middle Eastern	Baltimore, MD	*K pneumo*	Respiratory	PICU	VIM-4	CMY-4	Vent dep, Dandy-Walker	Kuwait	MEM, AMK	Died	[9]

Abbreviations: abx, antibiotics; ALL, acute lymphoblastic leukemia; AMK, amikacin; AML, acute myelogenous leukemia; CHD, congenital heart defect; COL, colistin; CSA, congenital skeletal abnormality; *E coli*, *Escherichia coli*; *E cloacae*, *Enterobacter cloacae*; ED, emergency department; FOS, fosfomycin; GNT, gentamicin; Heme-Onc, hematology-oncology ward; IMI, imipenem; *K pneumo*, *Klebsiella pneumoniae*; LEV, levofloxacin; MDS, myelodysplastic syndrome; MEM, meropenem; Multi, multiple; ND, no data; NICU, neonatal intensive care unit; NR, none reported; NT, no specific *Enterobacteriaceae* treatment; PC, personal communication; PICU, pediatric intensive care unit; RAD, reactive airway disease; Ref., reference; SBT, small bowel transplant; TIG, tigecycline; TMP/SMX, trimethoprim/sulfamethoxazole; Trach/Vent dep, tracheostomy/ventilator dependent; Unit, hospital location.

[a] If year isolated not available, year of publication reported.

[b] Presented to the ED; however, history of NICU stay overlapping with 2 other California *K pneumoniae* harboring bla_{IMP-4} NICU cases.

[c] L. K. Logan et al (unpublished data).

Targeted molecular surveillance can be cost effective and prevent unrecognized dissemination.

As in adults, the consequences of infection by MBL-producing GNB are likely to exact a disturbing toll. No one is safe.

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