Table 1. Susceptibility of *S. aureus* TCS mutants to various antibacterial agents

Strain	Inactivated gene		MIC (mg/L)							MBC (mg/L)	
	ORF no.[a]	Gene name[b]	OXA	VAN	TEC	BAC	GEN	ERY	OFX	hBD3	LL37
MW2	wild-type	—	8	2	0.5	64	4	0.25	1	8	4
FK61	MW0199	unassigned (HK)	8	2	0.5	64	4	0.25	1	8	4
FK62	MW0236	*lytS* (HK)	8	2	0.5	64	4	0.25	1	8	4
FK64	MW0621	*apsR* (RR)	4	2	0.5	64	1	0.25	1	4	2
FK65	MW0668	*saeR* (RR)	8	2	0.5	64	4	0.25	1	8	4
FK66	MW1208	unassigned (HK)	8	2	0.5	64	4	0.25	1	8	4
FK67	MW1305	*arlR* (RR)	2	2	0.5	64	2	0.25	1	2	1
FK68	MW1446	*srrA* (RR)	16	2	0.5	64	4	0.25	1	8	4
FK69	MW1637	*phoP* (RR)	8	2	0.5	64	4	0.25	1	8	4
FK71	MW1790	unassigned (HK)	8	2	0.5	64	4	0.25	1	8	4
FK72	MW1825	*vraS* (HK)	2	1	0.25	64	2	0.25	1	8	4
FK73	MW1962	*agrC* (HK)	8	2	0.5	64	4	0.25	1	8	4
FK74	MW2002	*kdpD* (HK)	8	2	0.5	64	4	0.25	1	8	4
FK75	MW2282	*hssR* (RR)	8	2	0.5	64	4	0.25	1	8	4
FK76	MW2314	*nreC* (HK)	8	2	0.5	64	4	0.25	1	8	4
FK77	MW2545	unassigned (HK)	8	2	0.5	16	4	0.25	1	8	4

OXA, oxacillin; VAN, vancomycin; TEC, teicoplanin; BAC, bacitracin; GEN, gentamicin; ERY, erythromycin; OFX, ofloxacin.
[a]Gene ID in *S. aureus* MW2.
[b]HK, histidine kinase; RR, response regulator.

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Transparency declarations

None to declare.

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Probable intrafamily transmission of a highly virulent CTX-M-3-producing *Escherichia coli* belonging to the emerging phylogenetic subgroup D_2 O102-ST405 clone

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Keywords: urinary tract infections, UTIs, extended-spectrum β-lactamases, ESBLs, uropathogenic strains

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Sir,

Over the last decade, an explosive spread of CTX-M-type extended-spectrum β-lactamases (ESBLs) in *Escherichia coli* has occurred, both in hospital and community settings. Two phenomena may explain such an epidemic profile: the spread within bacterial strains of the plasmids bearing the antibiotic resistance genes and the spread of bacterial clones bearing the resistance-encoding plasmids. The O25b-ST131 clone (where ST stands for sequence type), belonging to the B2 phylogenetic group, has disseminated all over the world.[1] Moreover, this clone, which is highly virulent, can capture a large panel of ESBL genes.[1–3] The emergence of such clones constitutes a major concern for public health. We report two members of the same family successively admitted to hospital for a febrile urinary tract infection (UTI) caused by a new emerging ESBL-producing *E. coli* clone.

Patient 1, a diabetic woman in her early 60s, was admitted to hospital for a pyelonephritis unsuccessfully treated with norfloxacin. She was obese and had a history of bariatric surgery 2 years ago. Therapy was switched to a combination of cefotaxime and amikacin. A urine culture taken at admission and screening for faecal carriage revealed an ESBL-positive *E. coli* resistant to ciprofloxacin and trimethoprim/sulfamethoxazole, but susceptible to cefoxitin, carbapenems, nitrofurantoin and aminoglycosides. The treatment was changed to ertapenem and the patient was managed by outpatient parenteral antimicrobial treatment for 2 weeks. She fully recovered and further urine cultures were sterile.

Patient 2, the son of Patient 1 who was in his early 40s, was admitted 10 days after for a febrile prostatitis. He was obese and had been admitted to another hospital 2 months before for the onset of type 2 diabetes. A urine culture and screening for faecal carriage identified an ESBL-positive *E. coli* with the same antibiotype pattern as the isolate from the mother. The patient was initially prescribed ofloxacin, but this was changed to ertapenem. The patient was discharged on day 6, with 3 weeks of home parenteral antibiotic therapy. He fully recovered and subsequent urine cultures were sterile. The son lived independently, but had regularly visited his mother for dinner during the preceding 6 months and had used her toilet. Neither patient had domestic pets.

A thorough analysis of one urine isolate and one faecal isolate from each patient was performed. The ESBL-producing isolates from the two patients were indistinguishable using enterobacterial repetitive intergenic consensus (ERIC)-1 and ERIC-2 PCR (Figure 1) and PFGE (data not shown) methods. The presence of $bla_{CTX\text{-}M\text{-}3}$ was revealed by PCR and sequencing.[4] Phylogenetic analysis by the PCR triplex method and multilocus sequence typing (MLST) using the Pasteur Institute scheme showed that these isolates belonged to the D_2 phylogenetic subgroup I (ST221), corresponding to ST405 of the Achtman MLST scheme.[4–6] The isolates exhibited the O102 type determined by allele-specific PCR, and harboured the virulence genes *aer*, *hlyC*, *iha*, *fuyA*, *traT*, *sat* and *papGII*, indicating the presence of the pathogenicity islands PAI I_{CFT073} and HPI.[2,4] These similarities indicated clearly that the four isolates belonged to the same clone. When tested in a mouse model of septicaemia, the mother's urine isolate was highly virulent, killing all 10 of the inoculated mice, as did a classical extraintestinal virulent strain of *E. coli*.[2]

Intrafamily transmissions of ESBL-positive *E. coli* responsible for UTIs are rarely described. A case of transmission between two family members of a CTX-M-15 ESBL-positive, *E. coli* O25b-ST131 strain was recently reported.[7] We describe here two members of the same family successively admitted to hospital for a febrile UTI due to a CTX-M-3 ESBL-positive *E. coli* strain belonging to the D_2 phylogenetic subgroup I-O102-ST405.

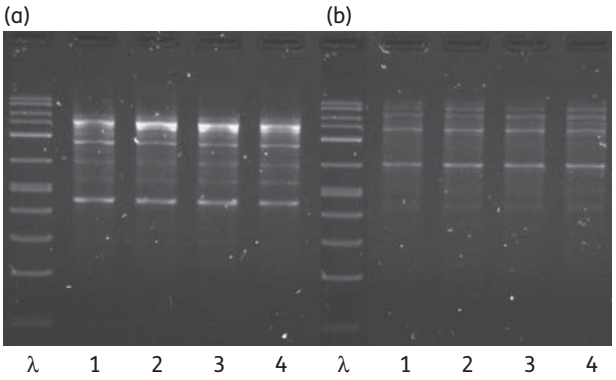

Figure 1. Typing of *E. coli* isolates using ERIC-1 PCR (a) and ERIC-2 PCR (b). λ, DNA molecular weight marker, Euroladder (Eurobio); lanes 1 and 2, *E. coli* isolates from urine and rectum of patient 1; lanes 3 and 4, *E. coli* isolates from urine and rectum of patient 2.

Potential mechanisms for the observed strain sharing include host-to-host transmission and acquisition from an external source, such as a food supply or domestic pets. We cannot rule out a common source as we did not investigate the presence of ESBL-positive *E. coli* in food, but the temporal pattern of their UTIs suggests a transmission from the mother to the son and the development of the infection from the commensal faecal reservoir. Neither patient had a history of prior antimicrobial use and, despite one hospitalization for each patient in the past 2 years, independent acquisition of an ESBL strain from distinct sources (especially of the same pulsotype) seems unlikely. Although both patients had diabetes, an underlying risk factor for complications of UTIs, neither of them had a previous history of febrile UTI. The strain appears to be particularly virulent, as inferred from two lines of evidence, namely the number and types of virulence genes (seven genes coding for adhesins, siderophores, toxins and protectins) and the mouse lethality assay. Recent studies indicate that the present D_2 O102-ST405 *E. coli* is an emerging clone, responsible, like the O25b-ST131 clone, for the worldwide spread of $bla_{CTX\text{-}M}$ genes.[8,9] This clone appears to pose the double threat of multidrug resistance and substantial extraintestinal virulence, in addition to its colonization and transmission ability. This makes its emergence and dissemination particularly concerning. New approaches to prevention, detection and management will be needed for this and similar clones.

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Transparency declarations

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Outbreak of *Klebsiella pneumoniae* producing KPC-2 and SHV-12 in a French hospital

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Keywords: carbapenemases, nosocomial infections, cross-transmission

Sir,

The emergence and dissemination of *Klebsiella pneumoniae* strains harbouring carbapenemases is a serious concern. *Klebsiella pneumoniae* carbapenemase (KPC) enzymes belong to molecular class A and are able to hydrolyse most β-lactams including carbapenems. Since the initial report of a KPC β-lactamase from a strain of *K. pneumoniae* in 1996, KPC producers have been reported from various geographical regions. Current reports indicate that KPC-producing isolates are widespread in China, Israel, Greece, South America and the USA, where the epidemiology of KPC in the hospital setting is changing.[1] Fortunately, these strains are still rare in western and northern Europe, but their detection remains difficult.[2]

Since 2003, patients hospitalized in the surgical ward of our hospital have been systematically screened on admission and weekly thereafter for intestinal carriage of bacteria producing extended-spectrum β-lactamases (ESBLs) and carbapenemases by plating rectal swabs on Drigalski agar containing 0.5 mg/L cefotaxime and MacConkey agar containing 2 mg/L ceftazidime (AES Laboratoire, Combourg, France). We report here four patients with *K. pneumoniae* producing KPC-2 and SHV-12. The first case was a patient transferred in July 2009 from Crete for treatment of recurrent angiocholitis on a biliary stent. The patient was negative on the day of admission, but 3 days later a further stool sample grew with ESBL-producing *K. pneumoniae*. This first isolate was not suspected to produce a carbapenemase since testing for susceptibility to imipenem using a disc diffusion method showed a diameter of 24 mm and an MIC of 1.5 mg/L by Etest (Bio-Rad, Marne la Coquette, France), both considered as susceptible according to the national recommendations of the Antibiogram Committee of the French Society for Microbiology.[3] However, in September, KPC-producing *K. pneumoniae* were isolated from three further patients (two from biliary fluid and one from tracheal fluid) hospitalized in the same ward at the same time. As KPC-producing *K. pneumoniae* are exceptional in France and described only in patients transferred from abroad (particularly Greece and Israel), it was decided to re-investigate all ESBL-producing *K. pneumoniae* isolated over the previous 6 months and to screen for carbapenemase production using the modified Hodge test and PCR. The only strain that was also positive for bla_{KPC} and which matched the three known KPC-positive isolates was the one isolated in July from the patient from Crete, who was thus potentially the index case. An epidemiological study (data not shown) revealed opportunities for cross-transmission to have occurred between the four patients.

This outbreak underlines the difficulty of identifying KPC-mediated carbapenem resistance using routine methods. The *K. pneumoniae* isolates from the latter three patients showed reduced susceptibility to imipenem, with a diameter of 21 mm, considered to indicate an intermediate level of resistance, an MIC of 2 mg/L and small colonies growing inside the zone of inhibition. All isolates, including the strain recovered from the Greek patient, exhibited resistance to other antibiotics tested: fluoroquinolones, tobramycin, amikacin and co-trimoxazole. The isolates were only susceptible to colistin and gentamicin. Using the modified Hodge test and EDTA-disc synergy,[4] all isolates were phenotypically positive for carbapenemase production, but negative for metallo-β-lactamase (MBL) production. ESBL screening tests using the double disc diffusion test between clavulanic acid and third-generation cephalosporins showed a synergy between ceftazidime or cefepime and clavulanic acid.

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