**Supplementary materials**

**Whole-genome sequencing, antimicrobial resistance genes, MLST, and phylogenetic analysis**

To determine the draft whole-genome sequence of the MBL-producing enterobacteriaceae isolates, DNA was extracted from the bacteria by phenol- chloroform treatment. We used a Nextera XT DNA library preparation kit (Illumina, Inc., CA, USA) to prepare DNA libraries for sequencing. Libraries were sequenced on a MiSeq system for 600 cycles (300-bp paired-end reads). Draft genomes (contigs) were obtained using the CLC genomic workbench (Qiagen) and SPAdes 3.9.1.[1](#_ENREF_1) Resfinder 2.1 was used for sequence analysis to determine antimicrobial-resistant genes in the isolated bacteria.[2](#_ENREF_2) Multilocus sequence typing (MLST) was performed using MLST 1.8 web tool (https://cge.cbs.dtu.dk//services/MLST/). Core genome single nucleotide polymorphism (SNP)-based phylogenetic analysis was performed with whole genome sequencing data. The MiSeq sequencing data were aligned to the genomic sequence of the reference isolate, *K. pneumoniae* DHQP1002001 ST34 (GenBank accession number CP016811.1) using the Burrows-Wheeler Aligner (BWA) with the “MEM” option.[3](#_ENREF_3) We constructed core-genome alignment using SAMtools (version 1.1) mpileup and VarScan (version 2.3.7) mpileup2cns and constructed a maximum-likelihood tree using RAxML.[4-6](#_ENREF_4)

**Plasmid analysis**

Plasmid incompatibility replicon typing was performed using PlasmidFinder.[7](#_ENREF_7) The complete nucleotide sequence of blaIMP-1-harboring IncN plasmid (pMTY14373\_IncN) contained different contigs generated using CLC Genomics Workbench and SPAdes. Plasmid multilocus sequence typing (pMLST) was performed using pMLST 1.4.[7](#_ENREF_7)

**Epidemiological investigation:**

Demographic and clinical data was following; age, sex, underlying diseases, the date of hospital admission, ward admission and discharge, the type of meals consumed, feeding assistance by nurses, radiographic studies, procedures, use of medical devices, administration of antibiotics and antacids, physiotherapy, surgery, echocardiograms, and radiographs. A case was defined as a patient with a positive culture for MBL-producing enterobacteriaceae, and the control was defined as a patient admitted to the same ward whose surveillance cultures remained negative during the outbreak.

**Accession number(s):**

The draft whole-genome sequence results of this study were deposited in the NCBI database under BioProject number AP018557. The draft genome sequences of 40 *K. pneumoniae* isolates were deposited in DDBJ and the NCBI BioSample database under accession numbers SAMD00114665 to SAMD00114701.

**Supplementary table**

|  |  |  |  |
| --- | --- | --- | --- |
| **Date of environmental surveillance culture** | Place from which culture sample was taken | Number of culture samples | Results of surveillance |
| May 23, 2014 | All sinks in the patients’ room, nurses’ station, bathroom, and sanitary room in the ward; ultrasound probe and lubricant | 28 | One sample from the sinks in the nurses’ station was positive. |
| June 6, 2014 | The ward’s lavatory bidet | 17 | Negative |
| June 13, 2014 | Sinks, milk bottle cabinet, computer keyboards, telephones, monitors, and refrigerator in the nurses’ station | 35 | Another sink at a nurses’ station near the milk bottle cabinet and the milk bottle cabinet itself were positive. |
| July 18, July | The keyboard of the ultrasound machines | 4 | Negative |
| August 8, 2014 | Frequently touched areas in the room such as the sink, electrical switches, suctioning devices, and doorknobs; doorknobs, lavatory seats, bidet, and safety rails in bathrooms throughout the ward | 76 | Negative |
| August 15, 2014 | All sinks throughout the ward | 22 | Negative |
| October 20, 2014 | The shower water and showerhead in adults’ and children’s bathrooms in the ward | 7 | Negative |
| December 18, 2014 | The tea, water in the tea dispenser, and the surfaces of the tea dispenser in the ward | 6 | Four samples from exit of two different nozzles and shaker in the tea dispenser were positive. |

**Keynote**

In total 195 samples were collected, and MBL-producing *K. pneumoniae* was isolated from seven environmental samples (3.6 %).

**Supplemental Figures**

**Figure 1.**



**Figure 1 legend**

Thirty-three (89.2 %) ST34 isolates of *Klebsiella pneumoniae* were isolated from stool (n=27), the tea dispenser (n=4), environment samples (n=1), and urine (n=1). Two isolates each of ST7 and ST359 were isolated from stool and environment samples, respectively.

Phylogenetic tree of *bla*IMP-1-positive ST34 strain of *K. pneumoniae* (n=33; TUM prefixes have been removed for clarity) constructed with maximum-likelihood phylogenetic analysis based on single nucleotide polymorphisms (SNPs) in the core genome. The core genome region was 89.68 % (5,047,282 / 5,628,146 bp) of the genome of the reference strain, *K. pneumoniae* DHQP1002001 ST34 (GenBank accession number CP016811.1). The scale distance corresponds to the number of substitutions per site. The circled number indicates the number of SNPs compared with *K. pneumoniae* TUM14373 as a representative common ancestor. No circled number indicates that there were no SNPs. An asterisk indicates that the organisms were isolated from the tea dispenser. The nucleotides to the left and right of the arrow indicate the nucleotide substitutions. Bold print indicates transversion mutations.

**Figure 2**



**Figure 2 legend**

A representative plasmid of IncN, pMTY14373\_IncN (GenBank accession number AP018557) was 51,787 bp in length, exhibited a GC content of 51.7%, and had 61 predicted ORFs. pMTY14373\_IncN belonged to pMLST-ST5 (Figure 2) and carried a class 1 integron containing *bla*IMP-1 [In798 with *aacA4’*-26, *bla*IMP-1 and *qacF*]. The nucleotide sequence of pMTY14373\_IncN highly resembled that of IncN pMLST-ST5 plasmid pKPI-6 (AB616660.2), which also carried a class 1 integron containing *bla*IMP-6 [*aac(6’)-Ib*, *bla*IMP-6, *aadA2*, and *tnp*] from *K. pneumoniae* (Figure 2). The pMTY14373\_IncN and pKPI-6 also harbored *bla*CTX-M-2 mediated by IS*Ecp1*

Comparison of pMTY14373\_IncN (GenBank accession number AP018557) carrying *bla*IMP-1 and *bla*CTX-M-2 with pKPI-6 (GenBank accession number AB616660.2) carrying *bla*IMP-6 and *bla*CTX-M-2 drawn with EasyFig, version 2.1. pMTY14373\_IncN and pKPI-6 were IncN plasmids belonging to pMLST-ST5. Block arrows indicate confirmed or putative open reading frames (ORFs) and their orientations. Arrow size is proportional to the predicted ORF length. The color code is as follows: green, replication initiation protein genes; blue, conjugal transfer genes; cyan, transposase genes; red, integrase genes; magenta, antibiotic resistance genes. Gray arrows represent putative, hypothetical, and unknown genes.

**References**

**1.** Bankevich A, Nurk S, Antipov D, et al. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. *J Comput Biol* 2012;19:455-477.

**2.** Yaita K, Aoki K, Suzuki T, et al. Epidemiology of extended-spectrum beta-lactamase producing Escherichia coli in the stools of returning Japanese travelers, and the risk factors for colonization. *PLoS One* 2014;9:e98000.

**3.** Li H, Durbin R. Fast and accurate short read alignment with Burrows-Wheeler transform. *Bioinformatics* 2009;25:1754-1760.

**4.** Li H, Handsaker B, Wysoker A, et al. The Sequence Alignment/Map format and SAMtools. *Bioinformatics* 2009;25:2078-2079.

**5.** Koboldt DC, Zhang Q, Larson DE, et al. VarScan 2: somatic mutation and copy number alteration discovery in cancer by exome sequencing. *Genome Res* 2012;22:568-576.

**6.** Stamatakis A. RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics* 2014;30:1312-1313.

**7.** Carattoli A, Zankari E, Garcia-Fernandez A, et al. In silico detection and typing of plasmids using PlasmidFinder and plasmid multilocus sequence typing. *Antimicrob Agents Chemother* 2014;58:3895-3903.