## Genome-Based Epidemiologic Analysis of VIM/IMP Carbapenemase-Producing *Enterobacter* spp., Poland

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We sequenced all nonduplicate 934 VIM/IMP carbapenemase-producing Enterobacterales (CPE) reported in Poland during 2006–2019 and found ≈40% of the isolates (n = 375) were *Enterobacter* spp. During the study period, incidence of those bacteria gradually grew in nearly the entire country. The major factor affecting the increase was clonal spread of several E. hormaechei lineages responsible for multiregional and interregional outbreaks (≈64% of all isolates), representing mainly the pandemic sequence type (ST) 90 or the internationally rare ST89 and ST121 clones. Three main VIM-encoding integron types efficiently disseminated across the clone variants (subclones) with various molecular platforms. Those variants were predominantly Pseudomonas aeruginosa-derived In238-like elements, present with IncHI2+HI2A, IncFII+FIA, IncFIB, or IncN3 plasmids, or chromosomal genomic islands in 30 Enterobacter STs. Another prevalent type, found in 34 STs, were In916-like elements, spreading in Europe recently with a lineage of IncA-like plasmids.

In the past few decades, bacterial infections with limited therapeutic options have become a serious threat for medicine. This problem is primarily caused by antimicrobial resistance (AMR), which disseminates by clonal spread of resistant organisms and horizontal transmission of mobile genetic elements with AMR genes. Several taxa have been classified as main AMR pathogens, including *Klebsiella pneumoniae* and *Enterobacter* spp. of the order Enterobacterales (1), and carbapenemase-producing Enterobacterales (CPE) are among the most challenging multidrug-resistant organisms (2). Important carbapenemase types, metallo- $\beta$ -lactamases (MBLs) of the families VIM and IMP, have been recorded in enterobacteria in Europe since 2001 (3), often in the Mediterranean region

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(4–10). The  $bla_{\text{VIM/IMP}}$  gene cassettes have usually been located in class 1 integrons, either assembled in *Pseudomonas* spp. and then transferred to Enterobacterales (4–6) or typical for Enterobacterales (4,7–10). The integrons have been carried by diverse plasmids with various replicons (4,7,8,10,11).

In Poland, VIM-type enzymes were originally identified in 2006 in *K. pneumoniae*, followed soon by *Enterobacter hormaechei* (12). Molecular analysis of all 121 VIM/IMP CPE isolates from 2006–2012 revealed high prevalence of *Enterobacter* spp. ( $\approx$ 53%) and relatively low contribution of *K. pneumoniae* ( $\approx$ 9%). *Enterobacter* spp. was dominated by *E. hormaechei* sequence type (ST) 90 and ST89, mostly with In238like integrons of *Pseudomonas aeruginosa* origin. We describe the genomic analysis of all VIM/IMP *Enterobacter* spp. isolates in Poland during 2006–2019, in the context of all VIM/IMP CPE from that period, and international *Enterobacter* spp. genomes from public databases.

#### Methods

## Study Design, Bacterial Isolates, Whole-Genome Sequencing, and Species Identification

The National Reference Centre for Susceptibility Testing conducts CPE surveillance in Poland, collecting isolates with basic patient, hospital ward, and isolate data. We tested the isolates by using CarbaNP (13) and phenotypic tests (14), and used PCRs for  $bla_{NDM}^-$ ,  $bla_{VIM}^-$ ,  $bla_{IMP}^-$ ,  $bla_{KPC}^-$ , and  $bla_{OXA-48}$ -like genes (4). A collection of 934 isolates from 246 hospitals in 117 cities were all nonduplicate VIM/IMP CPE confirmed during 2006–2019. We sequenced all those isolates by using MiSeq (Illumina, https://www.illumina.com), with de novo assemblies as described (15), and subjected them to species identification on the basis of average nucleotide identities by using FastANI 1.32 with a  $\geq$ 95%

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cutoff (16). We further analyzed the largest group of 375 isolates of the genus *Enterobacter* from 145 hospitals in 76 towns. We also sequenced 9 selected isolates by using MinION (Oxford Nanopore Technologies, https://nanoporetech.com) (15). We performed hybrid assemblies by using Unicycler 0.4.8 (17).

#### Molecular Typing and Comparative Genomic Analysis

We performed multilocus sequence typing (MLST) of all 375 Enterobacter spp. isolates (18) in silico by using mlst (https://github.com/tseemann/mlst). We performed the in-sample clonality single-nucleotide polymorphism (SNP) analysis for individual sequence types (STs) by using BioNumerics 7.6.3 (Applied Maths, https://www.applied-maths.com) and using index (i.e., initial) isolates of the STs as references. For the SNP-based phylogenetic analysis in the international context, we downloaded all (nonfiltered) 3,244 Enterobacter spp. genomes available in RefSeq (https://www.ncbi.nlm.nih.gov/refseq) as of June 6, 2022, and subjected them to MLST. We included isolates of the major STs (Appendix Table 1, https:// wwwnc.cdc.gov/EID/article/29/8/23-0199-App1. pdf) in our analysis, which we performed by using Parsnp 1.5.4 (https://github.com/marbl/parsnp). We visualized the Parsnp-generated phylotrees by using iTOL (https://itol.embl.de).

## Acquired AMR Genes, Integrons, and Plasmids or Genomic Islands Carrying *bla*vimme Genes

We detected acquired AMR genes by using ABRicate and the ResFinder database with 99.5% identity criterion (19) and profiled replicon types with Plasmid-Finder 2.1 (20). We performed structural analysis and annotation of MBL-encoding integrons, plasmids, and genomic islands manually in Geneious Prime 2022.0.1 (Biomatters, https://www.geneious.com) by using BLASTn (https://blast.ncbi.nlm.nih.gov/Blast. cgi). We visualized plasmid and island structures by using BRIG (http://brig.sourceforge.net) and Easyfig 2.2.5. (http://mjsull.github.io/Easyfig).

#### **Nucleotide Sequence Accession Numbers**

We submitted genomic data for the *Enterobacter* spp. isolates to the US National Center for Biotechnology Information (BioProject no. PRJNA877430). Plasmid sequences are available under the following Gen-Bank accession numbers: p743A, OQ111274; p5955A, OQ111275; p7753A, OQ111276; p4969H, OQ111277; p5435N, OQ111278; p5713F, OQ111279; p6234F, OQ111280. Sequences of genomic islands are available under the following GenBank accession numbers: *Eh*GI3, OQ116783; *Eh*GI4, OQ116782,.

#### Results

Taxonomic Distribution of VIM/IMP-Type CPE in Poland We collected 934 VIM/IMP CPE during 2006-2019 from 246 hospitals in 117 cities of all 16 regions of Poland (Appendix Figure 1, panel A). In annual numbers of cases, a gradual increase occurred, from a few cases during 2006-2008 up to 242 in 2019 (Appendix Table 2). We identified 9 genera, including Enterobacter (40.1%), Klebsiella (K. pneumoniae and K. oxytoca groups, 34.4%), Citrobacter (10.7%), Escherichia (9.2%), and Serratia (4.2%). The distribution of genera varied in time, including predominance of Enterobacter spp. and remarkable contribution of K. oxytoca during 2006–2013 (12) and still high prevalence of Enterobacter spp. but also a dynamic K. pneumoniae increase during 2014–2019 (Appendix Figure 1, panel B). Of note, annual numbers of Enterobacter spp. isolates grew at a roughly constant rate by the end of 2018, then escalating in 2019. VIM-type MBLs prevailed vastly (99.3%), whereas IMPs contributed marginally (0.7%). The 375 Enterobacter spp. isolates originated from 145 hospitals out of 76 towns and were recovered during various infections (64.3%), mainly of the urinary tract (31.5% of the infections) and wounds (28.6%), or from carriage (34.9%).

#### Species and Clonality of Enterobacter spp.

We identified 6 species among the 375 *Enterobacter* isolates, largely *E. hormaechei* (362 [96.5%]) with 5 subspecies: *steigerwaltii* (n = 244), *xiangfangensis* (n = 71), *hoffmannii* (n = 35), *oharae* (n = 11) and *hormaechei* (n = 1) (Appendix Table 2). The remaining species were *E. roggenkampii* (8 [2.2%]), *E. asburiae* (2 [0.5%]), and *E. kobei*, *E. ludwigii*, and *E. mori* (1 [0.3%] each). We distinguished 56 STs (Table, https://wwwnc.cdc.gov/ EID/article/29/8/23-0199-T1.htm; Figure 1); 5 STs had >10 isolates each (258 [68.8%]): ST90 (117 [31.2%] of all *Enterobacter* spp.), ST89 (74 [19.7%]), ST121 (36 [9.6%]), ST66 (18 [4.8%]), and ST134 (13 [3.5%]). Isolates of closely related STs (single-locus variants) represented clonal groups (CGs) or clonal complexes (CCs) (Table; Figure 1).

## $bla_{_{VIM}}$ and $bla_{_{IMP}}$ Genes and Their Integrons in *Enterobacter* spp.

We found 5  $bla_{\text{VIM}}$  genes, primarily of the  $bla_{\text{VIM-1}}$  group (91.5% of all MBLs in *Enterobacter* spp.); most were  $bla_{\text{VIM-4}}$  (49.1%),  $bla_{\text{VIM-1}}$  (40.6%), and  $bla_{\text{VIM-40}}$  (1.9%) (Table). The  $bla_{\text{VIM-2}}$  group included  $bla_{\text{VIM-20}}$  (4.0%) and  $bla_{\text{VIM-20}}$  (3.4%), whereas all  $bla_{\text{IMP}}$ s were  $bla_{\text{IMP-19}}$  (1.1%).

We characterized 16 integrons, including 4 new ones (Appendix Table 3). Elements of the In238 type

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**Figure 1.** Population structure of *Enterobacter* spp. isolates identified in a genome-based epidemiologic analysis of VIM/IMP carbapenemase-producing *Enterobacter* spp., Poland, 2006–2019. The minimum-spanning tree was constructed on the basis of 7-loci multilocus sequence type data. Each circle represents 1 ST, and each fragment of a pie chart corresponds to 1 isolate. The size of a circle is proportional to the number of isolates of that ST. Connecting lines infer phylogenetic relatedness in terms of several allelic differences (thick solid line indicates a single-locus variant, thin solid line indicates a double-locus variant). ST, sequence type.

prevailed (190 [50.4%]; 30 STs), carrying  $bla_{VIM4}$  (In238/ In238a)  $bla_{VIM40}$  (In1445), or  $bla_{VIM1}$  (In237a) genes. The second most prevalent In916 type (146 [38.7%]; 34 STs) had  $bla_{VIM1}$ . The  $bla_{VIM2}$ -like genes were located mostly in In1008-type integrons (26 [6.9%]; 5 STs), as  $bla_{VIM2}$ (In1008) or  $bla_{VIM20}$  (In1444).  $bla_{IMP19}$  was in a new element In2241. We noticed temporal changes in the integron distribution; the incidence of In238s grew from 2009 (n = 6) to 2014 (n = 24) and then stabilized, whereas that of In916 rapidly increased from the original identification in 2014 (n = 9) to 2019 (n = 57).

## Epidemiology of Major *E. hormaechei* Clones and Multiregional and Interregional Outbreaks

The most widespread clone was *E. hormaechei* subsp. *steigerwaltii* ST90 (117 [31.2%]), recorded during 2009–2019 in 58 hospitals in 38 cities, mostly in southern

regions (Figure 2; Appendix Table 4). Most of the 111 isolates with In238/In238a differed by 19–207 SNPs from the reference isolate (mean 71 SNPs) and formed a subclone (0–172 SNPs between closest relatives), likely resulting from multiregional expansion (outbreak I). We also classified 2 In238-carrying isolates of ST1762 (CC90) into this cluster (127–132 SNPs).

We observed *E. hormaechei* subsp. *steigerwaltii* ST89 (74 [19.7% of all isolates]) during 2006–2019 in 26 centers in 18 towns (Appendix Table 5, Figure 2). Most of the isolates (n = 67 [90.5% of ST89 isolates]) comprised 3 regional subclones with different integrons, representing outbreak II in Łódzkie (48 [0–75 SNPs between closest relatives]; In916), outbreak III in Wielkopolskie (12 [0–49 SNPs]; In1444), and outbreak IV in Kujawsko–Pomorskie (7 [4–12 SNPs]; In1445).

We identified *E. hormaechei* subsp. *xiangfangensis* CC121 isolates (ST121, 36 [9.6%]; ST1756, 3 [0.8%]) during 2014–2019 in 22 hospitals in 12 cities, mainly in the Mazowieckie and Łódzkie regions (Appendix Table 6, Figure 3). All those isolates were related to each other, with up to 84 SNPs with the reference (mean 46 SNPs); however, 2 outbreaks were distinguished based on the integron data: an interregional outbreak V (27 [0–46 SNPs between closest relatives]; In916) and a regional outbreak VI (6 [1–9 SNPs]; In238a).

Of the clones of lower incidence, ST66 and ST1754 (CG66; n = 19) were split into 2 genetically and geographically separated subclones (0–17 and 0–23 SNPs within the groups [404 SNPs between them]; both with In916), likely representing an interregional outbreak VII and a regional outbreak VIII (Appendix Table 7, Figure 4). ST134 (n = 13) showed variety as well, with a cluster of related organisms (9 [6–24 SNPs]; In238) arising from an apparent regional outbreak IX (Appendix Table 8, Figure 5).

## Phylogeny and International Context of Major *E. hormaechei* Clones

The clonal analysis of all 3,244 *Enterobacter* spp. genomes in RefSeq (as of June 6, 2022) revealed 546 STs; 61 STs were represented by >10 records. Out of the major VIM-positive clones in Poland, only ST90 and ST66 were among the 10 most numerous STs. Otherwise, the prevalent RefSeq clones were either not present (e.g., ST171 and ST133) or marginal (e.g., ST78 and ST114). However, the RefSeq genomes were unfiltered, which could have affected some of the observations. The phylogenetic analysis of 46 international ST90 genomes revealed 2 main clades and most of the 117 isolates in Poland, including outbreak I, belonged to a branch with several carbapenemase-free isolates from the



Figure 2. Geographic distribution and clonal analysis of Enterobacter hormaechei clonal complex 90 (ST90 and ST1762) in Poland, 2006-2019. A) Geographic distribution of the isolates; main administrative regions are labeled. Circles represent medical centers where the isolates were recorded. Sizes of the circles are proportional to numbers of cases of infection. B) SNP-based minimum-spanning tree of the isolates. Lengths of branches are related to numbers of SNPs between linked isolates. Numbers of SNPs are indicated above the branches or next to the dots. SNP, single nucleotide polymorphism; ST, sequence type.

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United Kingdom, France, Portugal, and Brazil (Appendix Figure 6).

ST89 was represented in RefSeq only by 2 isolates in Germany (1 with GIM-1) and 24 NDM-1-positive isolates in Poland during 2017-2020, which we analyzed in a previous study (21). Therefore, the phylotree comprised 100 isolates, including 98 from Poland (Appendix Figure 7), and consisted of 2 major lineages, each split then into multiple branches, correlating with the regional distribution of the isolates, regardless of their MBL content. The first lineage contained all of the VIM outbreak II isolates in Łódzkie plus a cluster of related NDM isolates from a neighboring area. The second lineage was divided into 2 major branches, 1 of which comprised the VIM outbreak IV in Kujawsko-Pomorskie and a large NDM epidemic from the adjacent region of Mazowieckie. The other branch contained mainly isolates from western Poland, including the VIM outbreak III from Wielkopolskie. Consistently, the 2 isolates in Germany were also located on the latter branch.

Only 7 ST121 genomes were present in RefSeq; the 36 VIM isolates in Poland, including outbreaks V– VI, formed 1 of 2 main lineages together with isolates from Brazil, Uganda, Morocco, Germany, and Poland (NDM) (21) (Appendix Figure 8). A total of 51 international ST66 isolates formed 2 lineages; 8 isolates in Poland of the outbreak VII belonged, primarily, to the lineage with isolates from Spain, France, and Germany mainly, whereas 10 outbreak VIII isolates clustered within the second lineage of more global character (Appendix Figure 9). ST134 records were sporadic in RefSeq (n = 9), and the 13 isolates in Poland, including outbreak IX, were located within 1 lineage together with single isolates from the United States, Lebanon, and Iran (Appendix Figure 10).

#### Resistomes

The resistome analysis demonstrated a large number and a variety of acquired AMR genes (6–27 genes per isolate; mean 15.8) (Appendix Table 9), in addition to the natural *Enterobacter* spp. *ampC* cephalosporinase genes. Their exact numbers could be specified only for the 9 MinION-sequenced genomes because some genes were in multiple copies in individual isolates (Appendix Table 10). The diversity of resistomes (AMR gene types and numbers) was common across and within the epidemic subclones; for some of those, the only stable AMR genes (i.e., present in all isolates of a subclone) were those in the MBL integrons. For instance, the ST90 isolates of the outbreak I had 67 AMR gene profiles, and ST89 isolates of the outbreak II had 33 AMR gene profiles. Along with *bla*<sub>VIM/IMP</sub>s, most of the isolates had genes coding for extended-spectrum  $\beta$ -lactamases ( $bla_{SHV}$  and  $bla_{CTX-M}$  types,  $bla_{GES-7'}$  and  $bla_{PER-2}$ ) or acquired AmpC-like cephalosporinases ( $bla_{CMY-83'}$   $bla_{DHA-1'}$  and  $bla_{FOX-20}$ ). Along with various aminoglycoside-modifying enzyme genes, numerous isolates had the 16S rRNA methylase gene *armA*, inactivating all aminoglycosides. Different variants of fluoroquinolone-resistance genes *qnrA*/*B*/*E*/*S* were commonly represented; 65 isolates contained the *mcr-9.1* colistin-resistance gene.

#### Plasmids Harboring *bla*<sub>VIM</sub> Genes

We identified 44 plasmid replicon types with 1–8 replicons per organism. The most frequent replicons were IncHI2 (n = 237), IncHI2A (n = 232), IncA (n = 165), IncFII (n = 140), and IncFIA (n = 116). Replicon profiles remarkably varied both between and within the subclones (Appendix Table 11). Long-read sequencing revealed the plasmid content, and the replicon and AMR gene distribution between the plasmids in 7 isolates representing the main epidemic subclones: ST90–In238 (n = 2; outbreak I), ST89–In916 (outbreak II), ST121–In916 (outbreak VI), ST121–In238a (outbreak IX) (Appendix Table 10). We performed the structural analysis on the plasmids with  $bla_{VIM}$ -harboring integrons.

In the 4 isolates with In238/In238a, including the 2 ST90-In238 representatives, the integrons were on 4 different plasmids. In 1 of those (isolate 4969-09), In238 was on an IncHI2+HI2A plasmid (p4969H; ≈261 kb), related to numerous others from Enterobacterales worldwide (91%–95% coverage;  $\approx 100\%$  identity), occasionally with *bla*<sub>IMP/VIM</sub> genes (Appendix Figure 11). One such plasmid from the Czech Republic, p51929\_MCR\_VIM (93% coverage; ≈100% identity), also contained In238 (22). The second ST90-In238 isolate (6234-09) had that integron on a plasmid with unique FII and FIA replicons (p6234F; ≈91 kb); FII was of some similarity to pECL\_A (≈83%) (23) and FIA to R27 (≈84%) (24). The IncFII+FIA scaffold matched 9 GenBank records well (>60% coverage, >98% identity) (Appendix Figure 12). Of note, in p4969H and p6234F, the In238 integron was located in novel, almost identical Tn21-like transposons Tn7536, similar to Tn1696 (25) (Appendix Figure 13).

The ST121–In238a isolate (5713–17) had In238a on an IncFIB-like plasmid (p5713F;  $\approx$ 120 kb), with the replicon similar to pB171 ( $\approx$ 91%) (26), homologous to 8  $bla_{VIM}$ -negative records (80%–90% coverage;  $\approx$ 100% identity) (Appendix Figure 14). Last, in the ST134–In238 isolate (5435–13) the integron resided on an IncN3-like plasmid (p5435N;  $\approx$ 46 kb), matching several records (89% coverage;  $\approx$ 100% identity), including

some with  $bla_{IMP/VIM}$  genes (Appendix Figure 15). The In238-type integrons in p5713F and p5435N were not located in Tn21-like transposons.

In the 3 isolates with In916: ST89 (7753–18), ST121 (743–14) and ST66 (5955–16), the integron resided on IncA plasmids (p7753A, ≈162 kb; p743A, ≈170 kb; and p5955A, ≈154 kb). Those isolates were highly related to each other and to 9 In916-carrying IncA plasmids (84%–96% coverage, ≈100% identity), including 5 from Italy (different *Enterobacterales*) (7) and 1 from Poland (*K. pneumoniae*) (27) (Appendix Figure 16). The plasmids varied mostly by rearrangements within the AMR region containing an IS26–bla<sub>SHV-12</sub>–In916–IS26 module (≈37.8-≈51.8 kb). This region in p743A was almost identical to plasmids pGB\_VIM and pGA\_VIM from Italy (7) (Appendix Figure 17).

#### Genomic Islands with blavim Genes

An isolate representing the epidemic subclone ST89–In1445 (8770–11; outbreak IV) had a new genomic island *Eh*GI3 with the *bla*<sub>VIM-40</sub> gene, and the isolate of the clone ST89–In1444 (2944–06; outbreak III) had another new genomic island with *bla*<sub>VIM-20</sub>. *Eh*GI3 (≈94.6 kb), inserted into the tRNA<sup>Gly</sup> gene, was a *clc*-like integrative and conjugative element (ICE) (41% coverage and ≈87% identity with the *clc* reference [28]), similar to ICEs found mainly in pseudomonads (29) (Appendix Figure 18). *Eh*GI4 (≈71.1 kb) was a mosaic region flanked by 2 IS26 copies with direct repeats, carrying In1444 and multiple AMR genes (e.g., *armA*).

#### Discussion

We describe VIM/IMP CPE in Poland, which markedly increased in recent years after a period of rather low prevalence. During 2017–2019, the annual VIM/ IMP CPE numbers recorded by the National Reference Centre for Susceptibility Testing (n = 545) were comparable with KPC (n = 686) or OXA-48 (n = 383) producers but far behind NDM organisms (n>6,000 [https://www.korld.nil.gov.pl]) (12,14,15,21,30). Among all carbapenemase-producing Enterobacter spp., the organisms with VIM/IMP-like enzymes were the predominant group (59.4%). The leading position of Enterobacter spp. among VIM/IMP CPE was maintained for all years of the study; however, the dynamic spread of *K. pneumoniae* in more recent years has notably changed the species composition. A substantial role of Enterobacter spp. among VIM CPE has been observed also in other countries of Europe (8,11).

The successful dissemination of VIM-producing *Enterobacter* spp. in Poland has depended largely on several epidemic subclones of *E. hormaechei* ST90,

ST89, and ST121 lineages, responsible for multiregional and interregional outbreaks I–VI ( $\approx$ 63% of all isolates). ST90 is a global clone, often reported with various carbapenemases (*11*). Its population in Poland has been dominated by the ST90–In238/In238a subclone, and since 2009 it has been expanding over a large territory (outbreak I). On the contrary, ST89 seems to be a local lineage, having been reported mostly in Poland with various VIMs, OXA-48, or NDM-1 so far. However, its repeated identification with GIM-1 in Germany indicates broader spread in central Europe (*21,31,32*).

The ST89 VIM-producing isolates in Poland were clustered into 3 regional subclones, ST89–In916, ST89–In1444, and ST89–In1445 (outbreaks II–IV), closely related to the previously described ST89 NDM-1 subclones from the same or neighboring areas (*21*). This finding indicates that ST89 has produced a series of regional sublineages, acquiring and then disseminating with different AMR genes. The epidemiology of ST121 has been unclear. According to RefSeq, it appears to be nonprevalent, although present broadly in the world. In Poland, it has spread extensively, acquiring several VIM integrons and causing major regional outbreaks (V-VI).

The second essential factor of the VIM-producing Enterobacter spp. expansion in Poland has been the horizontal transmission of 3 major VIM integron types. The In238 type with *bla*<sub>VIM-1</sub>-like genes and In1008 type with  $bla_{VIM,2}$ -like genes formed 2 evolving families of elements, with individual variants differing by mutations in *bla*<sub>VIM</sub> cassettes, and by 3'-termini of these in the case of In238 (specific 169bp repeats in some variants) (12,33). Both types were found originally in *P. aeruginosa* in Poland in 1998 (In238) (33) and 2001 (In1008) (34) and most likely were transmitted to Enterobacterales during 2006–2009 (12). However, In238 variants have been observed more broadly in central and southern Europe (22,35–37). The third major integron type, Enterobacterales-specific In916, has been recorded since the early 2010s in Spain, Italy, and France (4,8,11,38), and in Poland it has spread since at least 2013 (R. Izdebski and M. Gniadkowski, unpub. data). All those integron types have been acquired by E. hormaechei at the beginning of their dissemination in Enterobacterales in Poland with various molecular platforms.

In our previous study, the 2006–2012 predominant In238-type integrons in *E. hormaechei* ST89 and ST90 were assigned to IncHI2, PCR-nontypeable (largely), or IncM plasmids (12). We long-read sequenced 2 ST90–In238 isolates, representing outbreak I, in this study and found them to have In238

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on the IncHI2+HI2A or IncFII+FIA (previously nontypeable) plasmids, suggesting exchange between them. Given that the integron was located within almost identical Tn21-like transposons (Tn7536) in both plasmids, those might have been responsible for the inter-plasmid transfer. However, the 2 remaining long-read sequenced ST121 and ST134 isolates with In238/In238a had these integrons on yet other plasmids, IncFIB (ST121) and IncN3 (ST134), and not in a transposonic context. This finding indicates that acquisition and circulation of the In238-like elements among 36 STs of Enterobacter spp. in Poland have been multifactorial and complex phenomena. Regarding acquisition, an interesting case was provided by the ST89 isolate with the In238-like integron In1445, located within the *clc*-type ICE *Eh*GI3. In238 variants have been frequent in VIM-producing P. aeruginosa (39) and P. putida in Poland (40), being usually chromosomal in those. EhGI3 turned out to be almost identical to an ICE in 1 of the P. putida group isolates, indicating exchange of such elements between pseudomonads and Enterobacterales (P. Urbanowicz, M. Gniadkowski, unpub. data).

On the other hand, the proliferation of In916 seems to be relatively clear. In Europe, this integron has been associated with IncA, IncFII<sub>K</sub>, IncHI2, IncN, or PCR-nontypeable plasmids (4,7,8), and in our study isolates, it has entirely correlated with the IncA plasmids. A close relatedness between the In916-carrying IncA plasmids in Poland and Italy was proved, which together with high conjugative potential (7) have explained their spread on a large geographic scale. As in Italy (7) and France (8), rapid dissemination of these plasmids in Poland since 2013-2014 has contributed to the increase in VIM-producing Enterobacterales and Enterobacter spp., making In916 the most prevalent integron in 2019 (≈63%). The In916-carrying IncA plasmids occurred in 30 Enterobacter STs, including ST89, CC121 and CG66 subclones of 4 regional outbreaks, revealing that both the horizontal and clonal spread contributed to their recent proliferation.

Our study has shown the epidemiology of VIMproducing *Enterobacter* spp. during 14 years of VIM CPE surveillance in Poland, substantially updating the previous report (12). The results enable the precise definition of several *E. hormaechei* subclones of a remarkable epidemic potential, responsible for a series of territorial outbreaks, and enable the characterization of the main molecular platforms transmitting integrons with  $bla_{VIM}$  genes in *Enterobacter* populations. The study revealed several factors specific for Poland or central Europe, namely the prominent role of apparently rare *E. hormaechei* clones (ST89 or ST121), peculiar integrons of pseudomonadal origins (In238 and In1008 types), and unique VIM-encoding plasmids (IncFII+FIA with In238). We have also demonstrated some cosmopolitan elements, such as the global status of the epidemic ST90 clone and pan-Europe dissemination of In916-carrying IncA-like plasmids. All these observations indicate that AMR VIM-producing *E. hormaechei* and the VIM-encoding plasmids create an epidemiologic danger for hospital environments throughout Europe that clinicians and infection control specialists should be aware of.

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# etymologia revisited



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### Escherichia coli

[esh"a-rik'e-a co'lī]

Agram-negative, facultatively anaerobic rod, *Escherichia coli* was named for Theodor Escherich, a German-Austrian pediatrician. Escherich isolated a variety of bacteria from infant fecal samples by using his own anaerobic culture methods and Hans Christian Gram's new staining technique. Escherich originally named the common colon bacillus *Bacterium coli commune*. Castellani and Chalmers proposed the name *E. coli* in 1919, but it was not officially recognized until 1958.

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# Genome-Based Epidemiologic Analysis of VIM/IMP Carbapenemase-Producing *Enterobacter* spp., Poland

#### Appendix

Appendix Table 1. RefSeq assembly numbers, countries and STs of international *Enterobacter* spp. genomes used in phylogenetic analyses.

analyses.			
RefSeq assembly numbers	Country code	Country name	ST
GCF 000770745.2	AU	Australia	ST90
GCF_002416795.1	ZA	South Africa	ST90
GCF_002334585.1	JP	Japan	ST90
GCF_002237465_1	AU	Australia	ST90
GCF_002334485_1	IP	Janan	ST90
GCF_003175745_1	JP	Japan	ST90
CCE 002334605 1	IP	lanan	STOO
CCE_013403065_1	51 ER	France	STOO
CCE 020526705 1		Prozil	ST00
CCE_002022275_1		biazii	S190 ST00
GCF_002923275.1	JF	Japan Ore et Drite in	5190
GCF_900076435.1	GB	Great Britain	5190
GCF_900076635.1	GB	Great Britain	5190
GCF_900558595.1	GB	Great Britain	S190
GCF_001525015.1	AU	Australia	S190
GCF_001472575.1	GR	Greece	ST90
GCF_900077985.1	GB	Great Britain	ST90
GCF_018421195.1	DE	Germany	ST90
GCF_900076885.1	GB	Great Britain	ST90
GCF_013403105.1	FR	France	ST90
GCF 002334565.1	JP	Japan	ST90
GCF_002785805.1	CN	China	ST90
GCF_013403095.1	FR	France	ST90
GCF 900076465.1	GB	Great Britain	ST90
GCF_015209005_1	7A	South Africa	ST90
GCF_9000765551	GB	Great Britain	ST90
GCE 018447355 1	DE	Germany	ST90
GCE 002334545 1	IP	lanan	ST90
CCF_001475405_1	RO	Romania	STOO
$CCE_001653625.1$		Portugal	ST00
		Foltugal	5190
GCF_002334323.1	JP ZA	Japan South Africo	5190
GCF_015206945.1	ZA	South Africa	5190
GCF_900077715.1	GB	Great Britain	5190
GCF_002417315.1	ZA	South Africa	5190
GCF_900075615.1	GB	Great Britain	S190
GCF_900076495.1	GB	Great Britain	ST90
GCF_900536495.1	FR	France	ST90
GCF_900076825.1	GB	Great Britain	ST90
GCF_015208805.1	ZA	South Africa	ST90
GCF_002334505.1	JP	Japan	ST90
GCF_001526025.1	EC	Ecuador	ST90
GCF 900076765.1	GB	Great Britain	ST90
GCF_022551835.1	TW	Taiwan	ST90
GCF 900076525.1	GB	Great Britain	ST90
GCF_008931785.1	GB	Great Britain	ST90
GCF_002740875.1	RO	Romania	ST90
GCF 002510085.1	7A	South Africa	ST90
	<b></b> .		

RefSeq assembly numbers	Country code	Country name	ST
GCF_018420435.1	DE	Germany	ST89
GCF_900497145.1	DE	Germany	ST89
GCF_900076335.1	GB	Great Britain	ST66
GCF 022685505.1	ES	Spain	ST66
GCF_001524975.1	CA	Canada	ST66
GCF_015701095.1	US	USA	ST66
GCF_022685645.1	ES	Spain	ST66
GCF_003289405_1	FR	France	ST66
GCF_003289795_1	FR	France	ST66
CCE 016428335 1	ED	Franco	STEE
CCE_016428335.1		France	S100 ST66
		France	5100
GCF_022005705.1	ES	Spain	5100
GCF_013109535.1	0	Colombia	5166
GCF_012328865.1	FR	France	S166
GCF_022685625.1	ES	Spain	S166
GCF_009832255.1	US	USA	ST66
GCF_012328725.1	FR	France	ST66
GCF_013169525.1	CO	Colombia	ST66
GCF_022685745.1	ES	Spain	ST66
GCF 900076295.1	GB	Great Britain	ST66
GCF_019800365.1	DE	Germany	ST66
GCF_019837105.1	CN	China	ST66
GCF_012328705.1	FR	France	ST66
GCF 003289305 1	FR	France	ST66
GCF_022685685_1	FS	Spain	ST66
CCF_022685565_1	ES	Spain	ST66
CCE 020676025 1		Nigorio	5100
		Nigeria	5100
GCF_022005775.1	ES	Spain	5166
GCF_012328715.1	FR	France	5166
GCF_014190075.1	VN	Vietnam	5166
GCF_003977165.1	EG	Egypt	S166
GCF_016428365.1	FR	France	ST66
GCF_001473015.1	TW	Taiwan	ST66
GCF_900558375.1	GB	Great Britain	ST66
GCF_002208275.1	AU	Australia	ST66
GCF_002740595.1	TG	Togo	ST66
GCF 022685545.1	ES	Spain	ST66
GCF_004405115.1	CN	China	ST66
GCF_019448675.1	CN	China	ST66
GCF_900076615.1	GB	Great Britain	ST66
GCF_012328685.1	FR	France	ST66
GCF_012328885_1	FR	France	ST66
GCF_016428435_1	FR	France	ST66
GCF_019448715_1	CN	China	ST66
GCE 900076015 1	GB	Great Britain	ST66
CCE 00/181015 1	EP	Eranço	STEE
CCE 000538115 1	GR	Groat Britain	ST66
	GB		5100
GCF_021457515.1		Taiwali	5100
		Correction	3100 STCC
		Germany	5100 STCC
	ES	Spain	5100
	ES	Spain	5166
GCF_019/9/725.1	DE	Germany	5166
GCF_018422515.1	DE	Germany	ST121
GCF_002154935.1	BR	Brazil	ST121
GCF_023060195.1	BR	Brazil	ST121
GCF_020922915.2	PL	Poland	ST121
GCF_014842835.1	PK	Pakistan	ST121
GCF_001472215.1	MA	Morocco	ST121
GCF_003968685.1	UG	Uganda	ST121
GCF 014903845.1	US	ŪSA	ST134
GCF_000957655.1	US	USA	ST134
GCF_020567475.1	IR	Iran	ST134
GCF 900177445.1	-	-	ST134
GCE_900075455_1	GB	Great Britain	ST134
GCF_900447525.1	-	-	ST134
GCF 900075315 1	GR	Great Britain	ST134
GCF_011030345.1		Lebanon	ST134
GCF_003444755 1	CN	China	ST134
		Unina	01104

	groups/species/subs		Year													
genus	pecies	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	Total
Enterobacter	E. hormaechei	1	-	-	9	13	16	18	19	24	16	31	26	25	46	244
	subsp. <i>steigerwaltii</i>															
	E. hormaechei	-	-	-	-	2	-	-	-	6	5	2	16	14	26	71
	subsp.															
	xiangfangensis															
	E. hormaechei	-	-	-	-	-	-	1	-	3	2	5	5	5	14	35
	subsp. <i>hoffmannii</i>															
	E. hormaechei	-	-	-	-	-	-	1	-	-	7	-	2	-	1	11
	subsp. <i>oharae</i>															
	E. hormaechei	-	-	-	-	-	-	-	-	-	-	-	-	-	1	1
	subsp. hormaechei															
	E. roggenkampii	-	-	-	-	-	-	2	-	1	1	1	-	-	3	8
	E. asburiae	-	-	-	-	-	-	-	-	-	-	1	-	1	-	2
	E. kobei	-	-	-	-	-	1	-	-	-	-	-	-	-	-	1
	E. ludwigii	-	-	-	-	-	-	-	-	-	-	-	-	1	-	1
	E. mori	-	-	-	-	-	-	-	-	-	1	-	-	-	-	1
	Total	1	-	-	9	15	17	22	19	34	32	40	49	46	91	375
Klebsiella	K. pneumoniae	1	0	1	3	0	3	3	2	20	8	16	32	61	65	215
	group															
	K. oxytoca group				4	5	6	7	8	6	7	14	17	13	19	106
Citrobacter		-	-	-	-	-	1	1	3	9	6	8	14	29	29	100
Escherichia		-	-	-	-	-	-	-	2	7	4	11	17	13	32	86
Serratia		-	5	-	5	3	2	5	1	3	2	2	4	3	4	39
Morganella		-	-	-	-	-	-	-	-	-	1	1	1	3	1	7
Proteus		-	-	-	-	-	-	-	-	-	3	-	-	-	-	3
Leclercia		-	-	-	-	-	-	-	-	-	-	1	-	1	-	2
Hafnia		-	-	-	-	-	-	-	-	-	-	-	-	-	1	1
	Total	2	5	1	21	23	29	38	35	79	63	93	134	169	242	934

Appendix Table 2. Annual taxa distribution of VIM/IMP-producing Enterobacterales in Poland, 2006-2019

	0	ÿ	Number	Country, year and species of the first	
Integron type	Integron variant <sup>a</sup>	Gene cassette array	of STs	identification <sup>b, c</sup>	GenBank entry
with <i>bla</i> VIM-1-like gene	es				
ln238 (n=190)	In238 (n=160)	5'CS_aacA4_bla <sub>vIM-4rpt</sub> _3'CS	25	Poland, 1998, <i>P. aeruginosa</i>	AJ585042/AY702100 <sup>d</sup>
				Poland, 2008, <i>K. pneumoniae</i>	
				Poland, 2009, <i>E. hormaechei</i>	
	ln238a (n=22)	5'CS_aacA4_bla <sub>vIM-4</sub> _3'CS	9	Poland, 2009, <i>E. hormaechei</i>	JQ003906 (Hungary 2010)
	ln1445 (n=7)	5'CS_aacA4_bla <sub>VIM-40rpt</sub> _3'CS	1	Poland, 2011, <i>E. hormaechei</i>	MF678585
	In237a <sup>e</sup> (n=1)	5'CS_aacA4_bla <sub>VIM-1</sub> _3'CS	1	Poland, 2019, <i>K. oxytoca</i>	OQ116826
				Poland, 2019, <i>E. hormaechei</i>	
ln916 (n=146)	In916 (n=144)	5'CS_bla <sub>vIM-1</sub> _aacA4_aphA15_aadA1_catB2_3'CS	33	Spain, before 2014, <i>E. coli</i>	KF856617
				Poland, 2013, <i>E. coli</i> & <i>C. freundii</i>	
				Poland, 2014, <i>E. hormaechei</i>	
	<b>In2240</b> (n=2)	5'CS_bla <sub>vIM-1</sub> _aacA4_aphA15f_aadA1_catB2_3'CS	2	Poland, 2015, <i>E. hormaechei</i>	OQ116829
In70 (n=4)	In70	5'CS_bla <sub>VIM-1</sub> _aacA4_aphA15_aadA1_3'CS	2	Italy, 1997, <i>P. aeruginosa</i>	AJ969235
				Poland, 2011, <i>E. hormaechei</i>	
ln1654 (n=1)	In1654	5'CS_ <i>bla</i> vim-4rpt_3'CS	1	Poland, 2010, <i>P. aeruginosa</i>	MW595328
				Poland, 2014, <i>K. pneumoniae</i>	
				Poland, 2019, <i>E. hormaechei</i>	
ln110 (n=1)	In110	5'CS_ <i>bla<sub>VIM-1</sub>_aacA4_aadA1_</i> 3'CS	1	ltaly, 1999, <i>P. putida</i>	AJ439689
				Poland, 2006, <i>P. aeruginosa</i>	
				Poland, 2016, <i>E. hormaechei</i>	
In2238 (n=1)	In2238	5'CS_ <i>bla<sub>VIM-4rpt</sub>_bla<sub>OXA-2</sub>_aacA4_</i> 3'CS	1	Poland, 2016, <i>E. hormaechei</i>	OQ116827
In611 (n=1)	In611-like	5'CS_bla <sub>VIM-1</sub> _aacA4_aacC1_gcuP_gcuQ_∆aadA1	1	Poland, 2018, <i>E. hormaechei</i>	-
In2016 (n=1)	In2016-like <sup>f</sup>	5'CS_ <i>bla</i> <sub>VIM-4rpt</sub> _ <i>bla</i> <sub>OXA-10</sub> _∆3'CS	1	Poland, 2019, E. hormaechei	-
with <i>bla</i> vill-2-like gene	es				
In1008 (n=26)	In1008 (n=13)	5'CS blavim-2 aacA4 3'CS	4	Poland, 2001, <i>P. aeruginosa</i>	AM087408
				Poland, 2007, S. marcescens	
				Poland, 2009. E. hormaechei	
	In1444 (n=13)	5'CS blavim-20 aacA4 3'CS	2	Poland, 2006, E. hormaechei	MF678584
In2242 (n=2)	ln22`42 ´	5'CS_bla <sub>VIM-2</sub> _aacC11b_aadA6-10_3'CS	1	Poland, 2017, E. hormaechei	OQ116831
with <i>bla</i> IMP-like aenes	3				
In2241 (n=4)	In2241	5'CS_bla <sub>IMP-19</sub> _aacA4_aadA1b_catB2_3'CS	2	Poland, 2017, E. hormaechei	OQ116830

#### Appendix Table 3. VIM/IMP-encoding class 1 integrons identified in the Enterobacter study isolates

<sup>a</sup> - new integrons are indicated in bold style.

<sup>b</sup> – when the first report was from another country, then it is followed by the first Polish case(s); if the first Polish record was from non-Enterobacterales and/or non-Enterobacter Enterobacterales, it is then followed by the first Polish Enterobacterales and Enterobacter, respectively.

 <sup>a</sup> – date of isolation of the first Polish organism with a given integron may be earlier than that of the first isolate reported ever in another country.
 <sup>d</sup> – the original In238 record (AJ585042) contains a 2 nt error in the *bla*<sub>VIM4</sub> coding sequence; the subsequent *P. aeruginosa* In238 entry from Hungary from 2003 has been provided.
 <sup>e</sup> – In237a differs from the In237-like element, reported in Greece and Poland (GenBank acc. No. AY152821; Scoulica EV et al. Diagn Microbiol Infect Dis. 2004;48:167-72; Izdebski R et al J Antimicrob Chemother. 2018;73:2675-81), by having no 3'-terminal 169bp tandem repeat in the *blavim-1* gene cassette.

<sup>1</sup> - In2016-like differs from In2016, reported originally in P. aeruginosa in Poland (GenBank acc. No. MW595340; Urbanowicz P et al. J Antimicrob Chemother. 2021;76:2273-84), by having the 3'-terminal 169bp tandem repeat in the *bla*<sub>VIM-4</sub> gene cassette.

						Number of		
Isolate	Year	ST	Voivodeship	City	Hospital	SNPs	VIM variant	Remarks
4969-09 <sup>a</sup>	2009	ST90	Śląskie	Katowice	HS777	0	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
72-10	2010	ST90	Mazowieckie	Siedlce	HW222	19	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
865-13	2013	ST90	Dolnośląskie	Wrocław	WD4	19	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
5002-14	2014	ST90	Dolnośląskie	Legnica	HD1	20	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
93-10	2010	ST90	Dolnośląskie	Legnica	HD1	21	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6580-16	2016	ST90	Śląskie	Katowice	HS3	22	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6004-12	2012	ST90	Wielkopolskie	Poznań	HP1	23	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
1435-16	2016	ST90	Śląskie	Katowice	HS3	23	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
3728-12	2012	ST90	Mazowieckie	Otwock	HWA5	28	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
5917-12	2012	ST90	Świętokrzyskie	Skarżysko Kamienna	HT4	28	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
9482-18	2018	ST90	Śląskie	Katowice	HS3	29	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
9982-19	2019	ST90	Śląskie	Dąbrowa Górnicza	HS778	29	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6990-19	2019	ST90	Opolskie	Brzeg	HO3	32	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
2015-10	2010	ST90	Dolnośląskie	Wrocław	HD3	33	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
6360-12	2012	ST90	Mazowieckie	Radom	HM1	37	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
3103-13	2013	ST90	Mazowieckie	Warszawa	HW11	37	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
1847-19	2019	ST90	Świętokrzyskie	Końskie	HT7	38	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
3529-19	2019	ST90	Świętokrzyskie	Końskie	HT7	38	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
3601-19	2019	ST90	Świętokrzyskie	Końskie	HT7	38	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
8732-11	2011	ST90	Doľnoślaskie	Wrocław	HD3	39	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
2144-16	2016	ST90	Wielkopolskie	Poznań	HP1	39	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6340-16	2016	ST90	Ślaskie	Katowice	HS5	51	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
4210-15	2015	ST90	Ślaskie	Katowice	HS5	54	VIM-4	ST90-In238-VIM-4: multiregional outbreak I
4804-12	2012	ST90	Podkarpackie	Mielec	HR1	59	VIM-4	ST90-In238-VIM-4: multiregional outbreak I
723-15	2015	ST90	Małopolskie	Kraków	HK5	59	VIM-4	ST90-In238-VIM-4: multiregional outbreak I
6846-12	2012	ST90	Małopolskie	Kraków	HK5	60	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
5242-12	2012	ST90	Małopolskie	Kraków	HK3	61	VIM-4	ST90-In238-VIM-4: multiregional outbreak I
5923-13	2013	ST90	Małopolskie	Kraków	HK3	61	VIM-4	ST90-In238-VIM-4: multiregional outbreak I
1399-10	2010	ST90	Wielkopolskie	Poznań	HP1	62	VIM-4	ST90-In238-VIM-4: multiregional outbreak I
6294-17	2017	ST90	Małopolskie	Kraków	HK5	62	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6293-09	2009	ST90	Podkarpackie	Mielec	HR1	63	VIM-4	ST90-In238a-VIM-4: multiregional outbreak I
6438-12	2012	ST90	Ślaskie	Sosnowiec	AS1	63	VIM-4	ST90-In238-VIM-4: multiregional outbreak I
2794-19	2019	ST90	Małopolskie	Oświecim	HK667	64	VIM-4	ST90-In238-VIM-4: multiregional outbreak I
2977-15	2015	ST90	Małopolskie	Kraków	HK5	65	VIM-4	ST90-In238-VIM-4: multiregional outbreak I
5884-19	2019	ST90	Ślaskie	Katowice	HS26	65	VIM-4	ST90-In238-VIM-4: multiregional outbreak I
2224-10	2010	ST90	Wielkopolskie	Poznań	HP1	66	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
478-12	2012	ST90	Mazowieckie	Warszawa	HW1	66	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
2695-17	2017	ST90	Ślaskie	Bystra	HS16	66	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6017-09	2009	ST90	Podkarpackie	Mielec	HR1	67	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6335-09	2009	ST90	Małopolskie	Kraków	HK6	67	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6229-10	2010	ST90	Podkarpackie	Rzeszów	HR888	68	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
402-14	2014	ST90	Ślaskie	Chorzów	HS2	68	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
5237-14	2014	ST90	Lubelskie	Zamość	HL2	68	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
5375-14	2014	ST90	Lubelskie	Zamość	HL2	68	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
4034-10	2010	ST90	Lubuskie	Zielona Góra	HF2	68	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
2072-14	2014	ST90	Ślaskie	Sosnowiec	HS4	69	VIM-4	ST90-In238-VIM-4: multiregional outbreak I
216-15	2015	ST90	Lubelskie	Zamość	HL2	69	VIM-4	ST90-In238-VIM-4, multiregional outbreak I

Appendix Table 4. SNP scores between E. hormaechei subs. steigerwaltii CC90 (ST90 and ST1762) isolates<sup>a,b</sup>

						Number of		
Isolate	Year	ST	Voivodeship	City	Hospital	SNPs	VIM variant	Remarks
9300-11	2011	ST90	Lubelskie	Lublin	HL444	70	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
3571-13	2013	ST90	Małopolskie	Kraków	HK5	70	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
3845-15	2015	ST90	Podkarpackie	Krosno	HR7	70	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
5663-11	2011	ST90	Lubelskie	Zamość	HL2	71	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
4158-13	2013	ST90	Podkarpackie	Jasło	HR4	71	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
93-17	2017	ST90	Małopolskie	Kraków	HK8	71	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
4761-18	2018	ST90	Lubelskie	Zamość	HL2	71	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
9661-11	2011	ST90	Mazowieckie	Otwock	HWA3	72	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
9960-11	2011	ST90	Podkarpackie	Mielec	HR1	72	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6442-12	2012	ST90	Podkarpackie	Łańcut	HR3	72	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
4695-13	2013	ST90	Małopolskie	Kraków	HK5	72	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
814-14	2014	ST90	Lubelskie	Lublin	HL5	72	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
1152-14	2014	ST90	Lubelskie	Lublin	HL5	72	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
1167-15	2015	ST90	Małopolskie	Limanowa	HK7	72	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
584-17	2017	ST90	Świętokrzyskie	Starachowice	HT9	72	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
7995-18	2018	ST90	Małopolskie	Kraków	HK16	72	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6234-09	2009	ST90	Podkarpackie	Mielec	HR1	73	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
5310-10	2010	ST90	Podkarpackie	Rzeszów	HR888	73	VIM-4	ST90-In238a-VIM-4; multiregional outbreak I
9267-11	2011	ST90	Podkarpackie	Mielec	HR1	73	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
447-13	2013	ST90	Lubelskie	Lublin	HL5	74	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
2284-15	2015	ST90	Podkarpackie	Łańcut	HR3	74	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
4591-15	2015	ST90	Pomorskie	Gdańsk	HG1	74	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
3356-16	2016	ST90	Małopolskie	Kraków	HK5	74	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
2521-16	2016	ST90	Dolnośląskie	Polanica-Zdrój	HD11	74	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
444-11	2011	ST90	Podkarpackie	Mielec	HR1	75	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
1900-14	2014	ST90	Lubelskie	Puławy	HL6	75	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
875-17	2017	ST90	Małopolskie	Kraków	HK5	75	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
7625-18	2018	ST90	Podkarpackie	Jasło	HR4	76	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
4738-15	2015	ST90	Podkarpackie	Łańcut	HR3	77	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
7818-17	2017	ST90	Małopolskie	Limanowa	HK7	77	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
2218-14	2014	ST90	Podkarpackie	Jasło	AR2	78	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
218-17	2017	ST90	Podkarpackie	Rzeszów	HR5	78	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
5399-17	2017	ST90	Dolnośląskie	Wrocław	HD7	78	VIM-4	ST90-In238a-VIM-4; multiregional outbreak I
7398-17	2017	ST90	Małopolskie	Limanowa	HK7	78	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
7344-18	2018	ST90	Podkarpackie	Jasło	HR4	79	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
4148-13	2013	ST90	Śląskie	Chorzów	HS2	80	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
4149-13	2013	ST90	Śląskie	Chorzów	HS2	80	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
41-17	2016	ST90	Dolnośląskie	Polanica-Zdrój	HD11	80	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
4410-13	2013	ST90	Podkarpackie	Jasło	HR4	81	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
4537-13	2013	ST90	Śląskie	Chorzów	HS2	81	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
1000-11	2011	ST90	Podkarpackie	Mielec	HR1	82	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
4244-18	2018	ST90	Małopolskie	Andrychów	AK3	82	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
825-14	2014	ST90	Podkarpackie	Jasło	HR4	83	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
4928-11	2011	ST90	Małopolskie	Kraków	HK2	84	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
5072-10	2010	ST90	Świętokrzyskie	Kielce	HT555	85	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
5808-18	2018	ST90	Podkarpackie	Mielec	HR1	85	VIM-4	ST90-In238-VIM-4, multiregional outbreak I
7381-18	2018	ST90	Pomorskie	Wejherowo	HG7	85	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
7205-19	2019	ST90	Podkarpackie	Tarnobrzeg	HR889	85	VIM-4	ST90-In238-VIM-4; multiregional outbreak I

						Number of		
Isolate	Year	ST	Voivodeship	City	Hospital	SNPs	VIM variant	Remarks
3337-14	2014	ST90	Podkarpackie	Jasło	HR4	86	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
144-14	2013	ST90	Małopolskie	Kraków	HK2	89	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
3253-13	2013	ST90	Podkarpackie	Rzeszów	HR5	90	VIM-4	ST90-In238a-VIM-4; multiregional outbreak I
6747-18	2018	ST90	Lubelskie	Lublin	HL11	91	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6074-17	2017	ST90	Podkarpackie	Krosno	HR7	92	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6754-17	2017	ST90	Podkarpackie	Przemyśl	HR11	92	VIM-4	ST90-In238a-VIM-4; multiregional outbreak I
8600-19	2019	ST90	Śląskie	Siemianowice Śl.	HS1	93	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6492-18	2018	ST90	Małopolskie	Kraków	HK8	96	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6039-19	2019	ST90	Małopolskie	Kraków	HK5	115	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
3935-16	2016	ST90	Podkarpackie	Mielec	HR1	116	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
7910-19	2019	ST1762	Podkarpackie	Tarnobrzeg	HR890	127	VIM-4	ST1762-In238-VIM-4; multiregional outbreak I
3238-16	2016	ST1762	Podkarpackie	Nowa Dęba	HR8	132	VIM-4	ST1762-In238-VIM-4; multiregional outbreak I
5178-18	2018	ST90	Lubelskie	Lublin	HL14	133	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
6501-12	2012	ST90	Małopolskie	Bochnia	HK4	135	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
3526-14	2014	ST90	Mazowieckie	Radom	HM11	159	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
9518-19	2019	ST90	Małopolskie	Kraków	HK5	170	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
3525-14	2014	ST90	Mazowieckie	Radom	HM11	192	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
3524-14	2014	ST90	Mazowieckie	Radom	HM11	207	VIM-4	ST90-In238-VIM-4; multiregional outbreak I
10235-19	2019	ST90	Śląskie	Chorzów	HS2	649	VIM-1	ST90-In237a-VIM-1; single case
4083-09	2009	ST90	Mazowieckie	Warszawa	HW4	1631	VIM-4	ST90-In238-VIM-4; single case
3041-15	2015	ST90	Dolnośląskie	Wrocław	WD7	1653	VIM-1	ST90-In916-VIM-1; single case
2091-19	2019	ST90	Łódzkie	Łódź	HE7	1665	VIM-1	ST90-In916-VIM-1; single case
7154-17	2017	ST90	Dolnośląskie	Wrocław	WD7	1669	VIM-4+IMP-19	ST90-In238a-VIM-4-In2241-IMP-19; hospital
			•					dissemination
3723-18	2018	ST90	Dolnośląskie	Wrocław	WD7	1670	VIM-4+IMP-19	ST90-In238a-VIM-4-In2241-IMP-19; hospital
			•					dissemination

<sup>a</sup> – reference isolate, *i.e.* the Poland's index isolate of ST90/CC90 as confirmed by the National Reference Centre for Susceptibility Testing <sup>b</sup> – the SNP analysis of the CC90 isolates revealed 4092 polymorphic positions within ~4.0 Mb (77%) of the reference genome.

					Number of		
Isolate	Year	Voivodeship	City	Hospital	SNPs	VIM variant	Remarks
2944-06 <sup>a</sup>	2006	Lubuskie	Nowa Sól	HF20	0	VIM-20	ST89-In1444-VIM-20 regional outbreak III
5014-10	2010	Wielkopolskie	Poznań	HP2	28	VIM-20	ST89-In1444-VIM-20 regional outbreak III
258-11	2011	Wielkopolskie	Poznań	HP2	28	VIM-20	ST89-In1444-VIM-20 regional outbreak III
3033-13	2013	Wielkopolskie	Konin	HP7	28	VIM-20	ST89-In1444-VIM-20 regional outbreak III
5474-10	2010	Wielkopolskie	Poznań	HP2	30	VIM-20	ST89-In1444-VIM-20 regional outbreak III
9056-11	2011	Wielkopolskie	Poznań	HP100	30	VIM-20	ST89-In1444-VIM-20 regional outbreak III
2543-14	2014	Wielkopolskie	Gniezno	HP14	32	VIM-20	ST89-In1444-VIM-20 regional outbreak III
274-15	2015	Wielkopolskie	Piła	HP17	35	VIM-20	ST89-In1444-VIM-20 regional outbreak III
6630-12	2012	Wielkopolskie	Poznań	HP1	38	VIM-20	ST89-In1444-VIM-20 regional outbreak III
1064-16	2016	Wielkopolskie	Poznań	HP9	48	VIM-20	ST89-In1444-VIM-20 regional outbreak III
3584-16	2016	Lubuskie	Zielona Góra	AF1	49	VIM-20	ST89-In1444-VIM-20 regional outbreak III
195-11	2011	Mazowieckie	Grodzisk Mazowiecki	HW100	32	VIM-20	ST89-In1444-VIM-20 regional outbreak III
4884-09	2009	Dolnoślaskie	Wrocław	HD100	56	VIM-2	ST89-In1008-VIM-2 hospital outbreak
4885-09	2009	Dolnośląskie	Wrocław	HD100	56	VIM-2	ST89-In1008-VIM-2 hospital outbreak
8770-11	2011	Kujawsko-Pomorskie	Bydgoszcz	HC1	73	VIM-40	ST89-In1445-VIM-40 regional outbreak IV
5715-12	2012	Kujawsko-Pomorskie	Bydgoszcz	HC1	77	VIM-40	ST89-In1445-VIM-40 regional outbreak IV
3744-12	2012	Kujawsko-Pomorskie	Inowrocław	HC3	78	VIM-40	ST89-In1445-VIM-40 regional outbreak IV
533-12	2012	Kujawsko-Pomorskie	Bydgoszcz	HC1	80	VIM-40	ST89-In1445-VIM-40 regional outbreak IV
4261-12	2012	Kujawsko-Pomorskie	Inowrocław	HC3	80	VIM-40	ST89-In1445-VIM-40 regional outbreak IV
5863-12	2012	Kujawsko-Pomorskie	Inowrocław	HC3	85	VIM-40	ST89-In1445-VIM-40 regional outbreak IV
479-12	2012	Mazowieckie	Warszawa	HW9	77	VIM-40	ST89-In1445-VIM-40 regional outbreak IV
4642-14	2014	Mazowieckie	Mińsk Mazowiecki	HM2	52	VIM-4	ST89-In238-VIM-4 single case
43-16	2015	Mazowieckie	Siedlce	HM10	92	VIM-4	ST89-In2238-VIM-4 single case
1071-16	2016	Podlaskie	Białvstok	HB20	110	VIM-4	ST89-In238-VIM-4 single case
1100-16	2016	Lubuskie	Gorzów Wielkopolski	HF5	122	VIM-4	ST89-In238-VIM-4 single case
7753-18	2018	Łódzkie	Łódź	HE7	117	VIM-1	ST89-In916-VIM-1 regional outbreak II
9792-19	2019	Łódzkie	Łódź	HE7	117	VIM-1	ST89-In916-VIM-1 regional outbreak II
7654-19	2019	Łódzkie	Łódź	HE7	118	VIM-1	ST89-In916-VIM-1 regional outbreak II
7780-19	2019	Łódzkie	Łódź	HE112	119	VIM-1	ST89-In916-VIM-1 regional outbreak II
9398-18	2018	Łódzkie	Łódź	HE7	119	VIM-1	ST89-In916-VIM-1 regional outbreak II
6188-18	2018	Łódzkie	Łódź	HE7	120	VIM-1	ST89-In916-VIM-1 regional outbreak II
8201-18	2018	Łódzkie	Łódź	HE7	120	VIM-1	ST89-In916-VIM-1 regional outbreak II
3578-16	2016	Łódzkie	Łódź	HE7	126	VIM-1	ST89-In916-VIM-1 regional outbreak II
7339-17	2017	Łódzkie	Łódź	HE7	127	VIM-1	ST89-In916-VIM-1 regional outbreak II
8339-17	2017	Łódzkie	Łódź	HE7	127	VIM-1	ST89-In916-VIM-1 regional outbreak II
4253-16	2016	Łódzkie	Łódź	HE7	128	VIM-1	ST89-In916-VIM-1 regional outbreak II
4988-16	2016	Łódzkie	Łódź	HE7	129	VIM-1	ST89-In916-VIM-1 regional outbreak II
2917-18	2018	Łódzkie	Łódź	HE7	130	VIM-1	ST89-In916-VIM-1 regional outbreak II
7815-17	2017	Łódzkie	Łódź	HE7	130	VIM-1	ST89-In916-VIM-1 regional outbreak II
2284-18	2018	Łódzkie	Łódź	HE7	131	VIM-1	ST89-In916-VIM-1 regional outbreak II
6087-18	2018	Łódzkie	Łódź	HE111	131	VIM-1	ST89-In916-VIM-1 regional outbreak II
7133-16	2016	Łódzkie	Łódź	HE7	132	VIM-1	ST89-In916-VIM-1 regional outbreak II
7382-16	2016	Łódzkie	Łódź	HE7	132	VIM-1	ST89-In916-VIM-1 regional outbreak II
7338-17	2017	Łódzkie	Łódź	HE7	132	VIM-1	ST89-In916-VIM-1 regional outbreak II
5796-16	2016	Łódzkie	Łódź	HE7	133	VIM-1	ST89-In916-VIM-1 regional outbreak II
7019-16	2016	Łódzkie	Łódź	HE7	133	VIM-1	ST89-In916-VIM-1 regional outbreak II
1686-17	2017	Łódzkie	Łódź	HE7	133	VIM-1	ST89-In916-VIM-1 regional outbreak II

Appendix Table 5. SNP scores between E. hormaechei subs. steigerwaltii ST89 isolates<sup>a,b</sup>

					Number of		
Isolate	Year	Voivodeship	City	Hospital	SNPs	VIM variant	Remarks
7020-16	2016	Łódzkie	Łódź	HE7	134	VIM-1	ST89-In916-VIM-1 regional outbreak II
7383-16	2016	Łódzkie	Łódź	HE7	135	VIM-1	ST89-In916-VIM-1 regional outbreak II
4051-17	2017	Łódzkie	Łódź	HE17	135	VIM-1	ST89-In916-VIM-1 regional outbreak II
7517-19	2019	Łódzkie	Łódź	HE13	135	VIM-1	ST89-In916-VIM-1 regional outbreak II
6901-17	2017	Łódzkie	Łódź	HE7	136	VIM-1	ST89-In916-VIM-1 regional outbreak II
7381-16	2016	Łódzkie	Łódź	HE7	138	VIM-1	ST89-In916-VIM-1 regional outbreak II
1175-17	2017	Łódzkie	Łódź	HE7	138	VIM-1	ST89-In916-VIM-1 regional outbreak II
357-19	2019	Łódzkie	Łódź	HE7	140	VIM-1	ST89-In916-VIM-1 regional outbreak II
775-19	2019	Łódzkie	Łódź	HE7	140	VIM-1	ST89-In916-VIM-1 regional outbreak II
938-19	2019	Łódzkie	Łódź	HE7	140	VIM-1	ST89-In916-VIM-1 regional outbreak II
1008-19	2019	Łódzkie	Łódź	HE7	140	VIM-1	ST89-In916-VIM-1 regional outbreak II
1118-19	2019	Łódzkie	Łódź	HE7	140	VIM-1	ST89-In916-VIM-1 regional outbreak II
1888-19	2019	Łódzkie	Łódź	HE7	141	VIM-1	ST89-In916-VIM-1 regional outbreak II
7813-17	2017	Łódzkie	Łódź	HE7	141	VIM-1	ST89-In916-VIM-1 regional outbreak II
5105-19	2019	Łódzkie	Łódź	HE7	142	VIM-1	ST89-In916-VIM-1 regional outbreak II
5431-19	2019	Łódzkie	Łódź	HE7	142	VIM-1	ST89-In916-VIM-1 regional outbreak II
5741-19	2019	Łódzkie	Łódź	HE7	142	VIM-1	ST89-In916-VIM-1 regional outbreak II
9991-19	2019	Łódzkie	Łódź	HE7	142	VIM-1	ST89-In916-VIM-1 regional outbreak II
9312-19	2019	Łódzkie	Łódź	HE7	145	VIM-1	ST89-In916-VIM-1 regional outbreak II
9794-19	2019	Łódzkie	Łódź	HE7	150	VIM-1	ST89-In916-VIM-1 regional outbreak II
10251-19	2019	Łódzkie	Łódź	HE7	150	VIM-1	ST89-In916-VIM-1 regional outbreak II
8253-19	2019	Łódzkie	Radomsko	HE20	137	VIM-4	ST89-In1654-VIM-4 single case
6995-19	2019	Wielkopolskie	Poznań	HP11	131	VIM-1	ST89-In916-VIM-1 regional outbreak II
7480-19	2019	Wielkopolskie	Poznań	HP11	131	VIM-1	ST89-In916-VIM-1 regional outbreak II
5973-16	2016	Wielkopolskie	Poznań	HP11	132	VIM-1	ST89-In916-VIM-1 regional outbreak II
6126-18	2018	Wielkopolskie	Poznań	HP11	132	VIM-1	ST89-In916-VIM-1 regional outbreak II
5179-18	2018	Pomorskie	Wejherowo	HG7	122	VIM-1	ST89-In916-VIM-1 regional outbreak II

<sup>a</sup> - reference isolate, *i.e.* the Poland's index isolate of ST89 as confirmed by the National Reference Centre for Susceptibility Testing
 <sup>b</sup> - the SNP analysis of the ST89 isolates revealed 797 polymorphic positions within ~4.3 Mb (87%) of the reference genome.

						Number of	VIM	
Isolate	Year	ST	Voivodeship	City	Hospital	SNPs	variant	Remarks
743-14ª	2014	ST121	Mazowieckie	Warszawa	HW1	0	VIM-1	ST121-In916-VIM-1 interregional outbreak V
2876-15	2015	ST121	Mazowieckie	Warszawa	HW1	9	VIM-1	ST121-In916-VIM-1 interregional outbreak V
2902-15	2015	ST121	Mazowieckie	Otwock	HWA5	10	VIM-1	ST121-In916-VIM-1 interregional outbreak V
720-19	2019	ST121	Mazowieckie	Warszawa	HW9	30	VIM-1	ST121-In916-VIM-1 interregional outbreak V
4979-19	2019	ST121	Mazowieckie	Płońsk	HM4	30	VIM-1	ST121-In916-VIM-1 interregional outbreak V
2674-19	2019	ST121	Mazowieckie	Płońsk	HM4	31	VIM-1	ST121-In916-VIM-1 interregional outbreak V
5458-19	2019	ST121	Warmińsko-Mazurskie	Węgorzewo	HN9	31	VIM-1	ST121-In916-VIM-1 interregional outbreak V
6697-19	2019	ST121	Mazowieckie	Płońsk	HM4	31	VIM-1	ST121-In916-VIM-1 interregional outbreak V
7955-19	2019	ST121	Mazowieckie	Płońsk	HM4	31	VIM-1	ST121-In916-VIM-1 interregional outbreak V
3546-14	2014	ST121	Mazowieckie	Warszawa	HW1	34	VIM-1	ST121-In916-VIM-1 interregional outbreak V
2132-19	2019	ST121	Mazowieckie	Płońsk	HM4	36	VIM-1	ST121-In916-VIM-1 interregional outbreak V
188-16	2015	ST121	Mazowieckie	Warszawa	HW1	37	VIM-1	ST121-In916-VIM-1 interregional outbreak V
4600-15	2015	ST121	Mazowieckie	Warszawa	HW1	38	VIM-1	ST121-In916-VIM-1 interregional outbreak V
4220-14	2014	ST121	Łódzkie	Łódź	HE2	43	VIM-1	ST121-In916-VIM-1 interregional outbreak V
4728-14	2014	ST121	Łódzkie	Łódź	HE2	43	VIM-1	ST121-In916-VIM-1 interregional outbreak V
2977-14	2014	ST121	Dolnośląskie	Wrocław	HD999	44	VIM-1	ST121-In916-VIM-1 interregional outbreak V
4221-14	2014	ST121	Łódzkie	Łódź	HE2	44	VIM-1	ST121-In916-VIM-1 interregional outbreak V
8378-18	2018	ST121	Łódzkie	Łódź	HE7	50	VIM-1	ST121-In916-VIM-1 interregional outbreak V
209-18	2017	ST121	Podkarpackie	Przemyśl	HR11	56	VIM-1	ST121-In916-VIM-1 interregional outbreak V
7752-18	2018	ST1756	Łódzkie	Łódź	HE7	57	VIM-1	ST1756-In916-VIM-1 interregional outbreak V
4480-19	2019	ST1756	Łódzkie	Łódź	HE6	59	VIM-1	ST1756-In916-VIM-1 interregional outbreak V
1612-18	2018	ST121	Podkarpackie	Przemyśl	HR11	59	VIM-1	ST121-In916-VIM-1 interregional outbreak V
6350-18	2018	ST121	Podkarpackie	Przemyśl	HR11	64	VIM-1	ST121-In916-VIM-1 interregional outbreak V
5793-19	2019	ST1756	Łódzkie	Łódź	HE7	67	VIM-1	ST1756-In916-VIM-1 interregional outbreak V
7328-19	2019	ST121	Dolnośląskie	Wrocław	HD7	68	VIM-1	ST121-In916-VIM-1 interregional outbreak V
8602-19	2019	ST121	Podkarpackie	Rzeszów	HR17	68	VIM-1	ST121-In916-VIM-1 interregional outbreak V
5975-19	2019	ST121	Wielkopolskie	Ostrów Wielkopolski	HP18	70	VIM-1	ST121-In916-VIM-1 interregional outbreak V
7573-18	2018	ST121	Mazowieckie	Otwock	HWA2	34	VIM-4	ST121-In238a-VIM-4 regional outbreak VI
2589-19	2019	ST121	Mazowieckie	Otwock	HWA1	35	VIM-4	ST121-In238a-VIM-4 regional outbreak VI
3905-19	2019	ST121	Mazowieckie	Otwock	HWA1	37	VIM-4	ST121-In238a-VIM-4 regional outbreak VI
4784-19	2019	ST121	Mazowieckie	Warszawa	HW22	38	VIM-4	ST121-In238a-VIM-4 regional outbreak VI
5085-19	2019	ST121	Mazowieckie	Otwock	HWA3	40	VIM-4	ST121-In238a-VIM-4 regional outbreak VI
5713-17	2017	ST121	Mazowieckie	Warszawa	HW22	41	VIM-4	ST121-In238a-VIM-4 regional outbreak VI
7311-17	2017	ST121	Mazowieckie	Warszawa	HW21	70	VIM-2	ST121-In2242-VIM-2 regional dissemination
1969-19	2019	ST121	Mazowieckie	Konstancin-Jeziorna	HWA9	71	VIM-2	ST121-In2242-VIM-2 regional dissemination
1966-19	2019	ST121	Mazowieckie	Grójec	HM13	35	VIM-4	ST121-In238-VIM-4 single case
9390-18	2018	ST121	Mazowieckie	Warszawa	HW6	45	VIM-4	ST121-In238-VIM-4 single case
883-16	2016	ST121	Lubuskie	Gorzów Wielkopolski	HF5	75	VIM-4	ST121-In238-VIM-4 single case
6808-19	2019	ST121	Mazowieckie	Warszawa	HW567	84	VIM-4	ST121-In2016a-VIM-4 single case

Appendix Table 6. SNP scores between E. hormaechei subs. xiangfangensis CC121 (ST121 and ST1756) isolates<sup>a,b</sup>

<sup>a</sup> – reference isolate, *i.e.* the Poland's first index isolate of ST121/CC121 as confirmed by the National Reference Centre for Susceptibility Testing <sup>b</sup> – the SNP analysis of the CC121 isolates revealed 559 polymorphic positions within ~4.6 Mb (85%) of the reference genome.

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						Number of	VIM	
Isolate	Year	ST	Voivodeship	City	Hospital	SNPs	variant	Remarks
5955-16ª	2016	ST66	Mazowieckie	Majdan	HWA16	0	VIM-1	ST66-In916-VIM-1 interregional outbreak VII
1881-18	2018	ST66	Łódzkie	Łódź	HE13	10	VIM-1	ST66-In916-VIM-1 interregional outbreak VII
6334-17	2017	ST66	Łódzkie	Piotrków Trybunalski	HE19	13	VIM-1	ST66-In916-VIM-1 interregional outbreak VII
8063-17	2017	ST66	Łódzkie	Piotrków Trybunalski	HE19	13	VIM-1	ST66-In916-VIM-1 interregional outbreak VII
5530-19	2019	ST66	Mazowieckie	Majdan	HWA16	13	VIM-1	ST66-In916-VIM-1 interregional outbreak VII
2265-18	2018	ST66	Łódzkie	Zgierz	HE8	17	VIM-1	ST66-In916-VIM-1 interregional outbreak VII
1511-18	2018	ST66	Łódzkie	Łódź	HE11	18	VIM-1	ST66-In916-VIM-1 interregional outbreak VII
5642-17	2017	ST66	Łódzkie	Łódź	HE10	19	VIM-1	ST66-In916-VIM-1 interregional outbreak VII
5769-17	2017	ST66	Dolnośląskie	Wrocław	HD7	404	VIM-1	ST66-In916-VIM-1 regional outbreak VIII
7153-17	2017	ST66	Dolnośląskie	Wrocław	HD7	406	VIM-1	ST66-In916-VIM-1 regional outbreak VIII
7635-17	2017	ST66	Dolnośląskie	Wrocław	HD7	407	VIM-1	ST66-In916-VIM-1 regional outbreak VIII
5771-17	2017	ST66	Dolnośląskie	Wrocław	HD7	409	VIM-1	ST66-In916-VIM-1 regional outbreak VIII
6937-17	2017	ST66	Dolnośląskie	Wrocław	HD7	410	VIM-1	ST66-In916-VIM-1 regional outbreak VIII
5891-18	2018	ST66	Dolnośląskie	Wrocław	HD7	412	VIM-1	ST66-In916-VIM-1 regional outbreak VIII
9014-17	2017	ST66	Dolnośląskie	Wrocław	HD7	413	VIM-1	ST66-In916-VIM-1 regional outbreak VIII
9015-17	2017	ST66	Dolnośląskie	Wrocław	HD7	413	VIM-1	ST66-In916-VIM-1 regional outbreak VIII
6621-17	2017	ST66	Dolnośląskie	Wrocław	HD7	417	VIM-1	ST66-In916-VIM-1 regional outbreak VIII
6434-18	2018	ST1754	Dolnośląskie	Wrocław	HD101	425	VIM-1	ST1754-In916-VIM-1 regional outbreak VIII
6779-19	2019	ST66	Dolnośląskie	Wrocław	HD19	431	VIM-1	ST66-In916-VIM-1 regional outbreak VIII

Appendix Table 7. SNP scores between E. hormaechei subs. xiangfangensis CG66 (ST66 and ST1754) isolates<sup>a,b</sup>

<sup>a</sup> – reference isolate, *i.e.* the Poland's index isolate of ST66/CG66 as confirmed by the National Reference Centre for Susceptibility Testing

 $^{b}$  – the SNP analysis of the CG66 isolates revealed 511 polymorphic positions within ~4.5 Mb (88%) of the reference genome.

#### Appendix Table 8. SNP scores between E. hormaechei subs. steigerwaltii ST134 isolates<sup>a,b</sup>

					Number of		
Isolate	Year	Voivodeship	City	Hospital	SNPs	VIM variant	Remarks
5435-13ª	2013	Mazowieckie	Warszawa	HW1	0	VIM-4	ST134-In238-VIM-4 regional outbreak IX
5436-13	2013	Mazowieckie	Warszawa	HW1	7	VIM-4	ST134-In238-VIM-4 regional outbreak IX
2118-14	2014	Mazowieckie	Warszawa	HW1	14	VIM-4	ST134-In238-VIM-4 regional outbreak IX
1958-15	2015	Mazowieckie	Warszawa	HW9	25	VIM-4	ST134-In238-VIM-4 regional outbreak IX
6302-16	2016	Mazowieckie	Warszawa	HW24	29	VIM-4	ST134-In238-VIM-4 regional outbreak IX
5897-16	2016	Mazowieckie	Warszawa	HW6	30	VIM-4	ST134-In238-VIM-4 regional outbreak IX
1258-16	2016	Mazowieckie	Warszawa	HW24	30	VIM-4	ST134-In238-VIM-4 regional outbreak IX
885-16	2016	Mazowieckie	Warszawa	HW24	31	VIM-4	ST134-In238-VIM-4 regional outbreak IX
151-17	2017	Mazowieckie	Warszawa	HW24	31	VIM-4	ST134-In238-VIM-4 regional outbreak IX
2791-19	2019	Świętokrzyskie	Kielce	HT6	192	VIM-1	ST134-In916-VIM-1 hospital dissemination
3340-19	2019	Świętokrzyskie	Kielce	HT6	193	VIM-1	ST134-In916-VIM-1 hospital dissemination
4986-14	2014	Dolnośląskie	Wrocław	HD4	208	VIM-4	ST134-In238a-VIM-4 hospital dissemination
752-15	2015	Dolnośląskie	Wrocław	HD4	211	VIM-4	ST134-In238a-VIM-4 hospital dissemination

<sup>a</sup> – reference isolate, *i.e.* the Poland's index isolate of ST134 as confirmed by the National Reference Centre for Susceptibility Testing

<sup>b</sup> - the SNP analysis of the ST134 isolates revealed 307 polymorphic positions within ~4.5 Mb (89%) of the reference genome.

#### Appendix Table 9. Resistomes of the Enterobacter spp. isolates

<u>- appondix</u>					acquired AMR genes <sup>a</sup>		n AMR
taxa/ST	integron	n isolates	remarks	resistance to β-lactams	resistance to aminoglycosides	resistance to other groups	genes/ isolate
E. normaec ST45	<i>chei</i> subsp. In238	steigerwaltii 7	i	bla <sub>VIM-4</sub> , (bla <sub>CTX-M-3</sub> ), (bla <sub>CMY-83</sub> ), bla <sub>SHV-</sub> 5	(aac(3)-la), (aac(6')-lm), aac(6')-lb, (aadA1), (aph(2'')-lla), (aph(3'')-lb), aph(6)-	(dfrA1), dfrA14, sul1, (tet(A)), tet(B), (catA1), (catA2), oqxB	12.3
	In916	2		bla <sub>VIM-1</sub> , (bla <sub>CTX-M-9</sub> ), (bla <sub>SHV-12</sub> ), bla <sub>TEM-</sub>	aac(6')-lb, aadA1, (aadA2), (ant(2'')-la), apb(3'')-lb, apb(6)-ld, apb(3')-XV	dfrA14, sul1, sul2, (qnrA1), (qnrS1), (tet(A)), catA2, catB2, ogxB, (mcr-9,1)	18.0
ST62	In916	1		bla <sub>VIM-1</sub> , bla <sub>SHV-12</sub> , bla <sub>LAP-2</sub>	aac(6')-lb, aadA1, aadA16, aph(3')-lb, aph(6)-ld, aph(3')-XV	dfrA14, dfrA27, sul1, sul2, qnrS1, fosA, tet(A), catB2, oqxB, arr	19.0
ST89	In916	48	outbreak II	bla <sub>VIM-1</sub> , (bla <sub>CTX-M-3</sub> ), (bla <sub>CTX-M-15</sub> ), (bla <sub>CTX-M-256</sub> ), <sup>b</sup> (bla <sub>SHV-12</sub> ), (bla <sub>OXA-1</sub> ), (bla <sub>TEM-1</sub> )	(aac(3)-IId), (aac(3)-IIe), aac(6')-Ib, (aac(6')-Ib-cr), aadA1, (aph(3'')-Ib), (aph(6)- Id), aph(3')-XV	(sat2), (dfrA1), (dfrA14), (sul1), (sul2), (qnrS1), fosA, (tet(A)), (catA1), catB2, (catB3), (oqxA), (oqxB), (mphA)	19.6
	In238	3		bla <sub>VIM-4</sub> , (bla <sub>CTX-M-3</sub> ), (bla <sub>SHV-12</sub> ), (bla <sub>TEM-1</sub> )	(aac(3)-IIe), aac(6')-Ib, (aadA2), (aph(3')- VIa), (aph(3'')-Ib), (aph(6)-Id), armA	(sat2), (dfrA1), (dfrA12), (dfrA19), (sul1), (sul2), fosA, (tet(D)), mphE, msrE, oqxB, (mcr-9.1)	14.7
	In1654	1		bla <sub>VIM-4</sub> , bla <sub>CTX-M-3</sub> , bla <sub>OXA-1</sub>	aadA2, ant(2")-la, armA	sat2, dfrA1, dfrA12, sul1, qnrS1, fosA, tet(A), mphE, msrE	15.0
	In2238	1		bla <sub>VIM-4</sub> , bla <sub>CTX-M-3</sub> , bla <sub>OXA-2</sub> , bla <sub>TEM-1</sub>	aac(6')-lb, aadA1, aadA2, armA	dfrA1, dfrA12, sul1, fosA, catA1, msrE, oaxB	15.0
	In1445	7	outbreak IV	bla <sub>VIM-40</sub> , bla <sub>CTX-M-3</sub>	aac(6')-lb, (aph(3')-VI), aph(3'')-lb, aph(6)- Id	sul1, fosA, oqxB	9.8
	In1008	2		bla <sub>VIM-2</sub> , bla <sub>CTX-M-3</sub> , bla <sub>SHV-5</sub>	aac(6')-lb, aadA2, armA	dfrA12, (sul1), fosA, (tet(A)), mphE, msrE, (oaxB)	12.5
	In1444	12	outbreak III	bla <sub>VIM-20</sub> , (bla <sub>CTX-M-3</sub> )	(aac(3)-IId), aac(6')-Ib, (aadA2), (aph(3')- Ia), (armA)	(dfrA12), sul1, fosA, (mphE), (msrE), oaxB	12.6
ST90	In238	107	outbreak I	bla <sub>VIM-4</sub> , (bla <sub>CTX-M-3</sub> ), (bla <sub>CTX-M-15</sub> ), (bla <sub>DHA-1</sub> ), (bla <sub>SHV-5</sub> ), (bla <sub>SHV-12</sub> ), (bla <sub>OXA-1</sub> ), (bla <sub>OXA-10</sub> ), (bla <sub>TEM-1</sub> ), (bla <sub>LAP-2</sub> )	(aac(3)-IIa), (aac(3)-IId), (aac(3)-IIe), (aac(6')-IIc), aac(6')-Ib, (aac(6')-Ib-cr), (aadA1), (aadA2), (ant(2")-Ia), (aph(2")-IIa), (aph(3')-Ia), (aph(3")-Ib), (aph(6)-Id), (armA)	(dfrA1), (dfrA12), (dfrA14), (dfrA19), (sul1), (sul2), (qnrA1), (qnrB1), (qnrB4), (qnrS1), (fosA), (tet(A)), (tet(D)), (catA2), (catB3), (cmlA1), (mphE), (msrE), (oqxB) (mcr-9 1) (arr)	14.6
	In238a	5		bla <sub>vIM-4</sub> , bla <sub>CTX-M-3</sub> , (bla <sub>SHV-12</sub> ), (bla <sub>TEM-1</sub> ), (bla <sub>TEM-1</sub> )	(aac(3)-II), (aac(3)-IIe), aac(6')-Ib, (aac(6')- IIc), (aadA1), (aadA2), (aph(3'')-Ib), (armA)	(dfrA1), (dfrA12), (dfrA19), sul1, (sul2), (qnrA1), fosA, (tet(D)), (mphE), (msrE), (mcr-9.1), (arr)	12.8
ST1762	In238	2		bla <sub>VIM-4</sub> , bla <sub>CTX-M-3</sub> , bla <sub>TEM-1</sub>	aac(3)-IId, aac(6')-Ib, aadA1, aadA2, aph(3'')-Ib, aph(6)-Id, armA	(dfrA1), (dfrA12), dfrA19, sul1, sul2, qnrA1, fosA, tet(D), catA2, mphE, msrE, mcr-9.1	22.0
ST90	In238a+ <b>In2241</b>	2		bla <sub>VIM-4</sub> , bla <sub>IMP-19</sub> , bla <sub>LAP-2</sub> , bla <sub>TEM-1</sub>	aac(3)-II, aac(6')-Ib, aadA1b, aadA2, (aph(3')-Ia), aph(3'')-Ib, aph(6)-Id	dfrA19, sul1, fosA, qnrS1, catB2, arr	17.5
	In916	1		bla <sub>VIM-1</sub> , bla <sub>CTX-M-15</sub> , bla <sub>SHV-12</sub> , bla <sub>OXA-1</sub> , bla <sub>TEM-1</sub>	aac(3)-Ile, aac(6')-Ib, aadA1, aph(3')-XV	dfrA14, sul1, sul2, qnrB1, qnrS1,tet(A), fosA, catB2	17.0
	In2240	1		bla <sub>viM-1</sub>	aac(6')-lb, aadA1, aph(3'')-lb, aph(6)-ld, aph(3')-XV	sul1, sul2, qnrS1, fosA, catB2	11.0
	In237a	1		<i>bla</i> <sub>VIM-1</sub> , <i>bla</i> <sub>OXA-1</sub> , <i>bla</i> <sub>TEM-1</sub>	aac(3)-IId, aac(6')-Ib, aph(3")-Ib, aph(6)-Id	dfrA14, sul1, sul2, qnrB1, fosA, tet(D), mphE, msrE	15.0
ST91	In1008	2		bla <sub>VIM-2</sub> , bla <sub>OXA-1</sub> , bla <sub>TEM-1</sub>	aac(3)-IId, aac(6')-Ib, aadA1, aph(3")-Ib, aph(6)-Id, armA	dfrA14, sul1, sul2, qnrB1, fosA, tet(A)	16.0

					acquired AMR genes <sup>a</sup>		n AMR
taxa/ST	intearon	n isolates	remarks	resistance to β-lactams	resistance to aminoglycosides	resistance to other groups	genes/ isolate
	In70	3		bla <sub>VIM-1</sub> , (bla <sub>SHV-12</sub> ), (bla <sub>LAP-2</sub> )	aac(6')-lb, (aac(6')-lb-cr), aadA1, (aadA2), (aadA16), ant(2")-la, (aph(3')-la), aph(3')- Vla, aph(3")-lb, aph(6)-ld, aph(3')-XV	(dfrA14), (dfrA27), sul1, sul2, (qnrB6), qnrS1, fosA, tet(A), (arr-3), catB2, (floR)	21.0
	In916	1		bla <sub>VIM-1</sub> , bla <sub>CTX-M-15</sub> , bla <sub>OXA-1</sub> , bla <sub>SHV-12</sub>	aac(3)-IIe , aac(6')-Ib, aadA1, ant(2'')-Ia, aph(3')-VIa, aph(3'')-Ib, aph(6)-Id , aph(3')- XV	dfrA14, sul1, sul2, qnrB1, qnrS1, fosA, tet(A), catB2	20.0
ST93	In238	2		$bla_{VIM-4}, bla_{SHV-5}, (bla_{LAP-2}), (bla_{TEM-1})$	aac(3)-la, aac(6')-lb, (aadA1)	(dfrA1), sul1, (qnrS1), fosA, (tet(B)), (tet(G)), (catA1), (floR2), oaxB	12.5
	In1008	1		bla <sub>VIM-2</sub> , bla <sub>CTX-M-3</sub> , bla <sub>TEM-1</sub>	aac(3)-IId, aac(6')-Ib, armA	sul1, fosA, oqxB	9.0
ST106	In916	3		bla <sub>VIM-1</sub> , bla <sub>CTX-M-15</sub> , bla <sub>OXA-1</sub> , bla <sub>TEM-1</sub>	aac(6')-lb, (aac(6)-ld), aadA1, aph(3')-XV	dfrA14, sul1, sul2, qnrB1, fosA, tet(A), catB2, (oqxB)	16.0
ST110	In916	1		bla <sub>vIM-1</sub>	aac(6')-Ib, aadA1, aph(3")-Ib, aph(6)-Id, aph(3')-XV	sul1, sul2, qnrS1, fosA, catB2, oqxB	12.0
ST116	In2240	1		bla <sub>∨IM-1</sub>	aac(6')-Ib, aadA1, aph(3'')-Ib, aph(6)-Id, aph(3')-XV	sul1, sul2, qnrS1, fosA, catB2	11.0
ST134	In238	9	outbreak IX	bla <sub>VIM-4</sub> , bla <sub>CTX-M-15</sub> , bla <sub>OXA-1</sub>	aac(3)-lle, aac(6')-lb, aac(6')-lb-cr, aadA1	dfrA14, (sul1), qnrB1, (catA1), fosA, tet(A), oqxB	14.0
	In238a	2		bla <sub>VIM-4</sub> , bla <sub>CTX-M-15</sub> , bla <sub>OXA-1</sub> , bla <sub>TEM-1</sub>	aac(3)-IIe, aac(6')-Ib, aac(6')-Ib-cr, aadA1, aph(3'')-Ib, aph(6)-Id	(dfrA12), (dfrA14), sul1, sul2, qnrB1, fosA, tet(A), oqxB	18.0
	In916	2		<i>bla</i> <sub>VIM-1</sub> , <i>bla</i> <sub>CTX-M-15</sub> , <i>bla</i> <sub>SHV-12</sub> , <i>bla</i> <sub>OXA-1</sub> , <i>bla</i> <sub>TEM-1</sub>	aac(3)-IIe, aac(6')-Ib, aadA1, aph(3')-XV	dfrA14, sul1, sul2, qnrB1, qnrS1, fosA, tet(A), catB2	18.0
ST175	In238a	2		bla <sub>VIM-4</sub> , bla <sub>CTX-M-15</sub> , bla <sub>OXA-1</sub> , bla <sub>TEM-1</sub>	aac(3)-IIe, aac(6')-Ib, aac(6')-Ib-cr, aadA1, aph(3'')-Ib, aph(6)-Id	dfrA14, sul1, sul2, qnrB1, fosA, tet(A)	17.0
	In916	1		<i>bla</i> <sub>VIM-1</sub> , <i>bla</i> <sub>CTX-M-15</sub> , <i>bla</i> <sub>SHV-12</sub> , <i>bla</i> <sub>OXA-1</sub> , <i>bla</i> <sub>TEM-1</sub>	aac(3)-IIe, aac(6')-Ib, aadA1, aph(3'')-Ib, aph(3')-XV	dfrA14, sul1, sul2, qnrB1, qnrS1, fosA, tet(A), catB2	18.0
ST184	In916	1		bla <sub>VIM-1</sub> , bla <sub>GES-7</sub> , bla <sub>PER-2</sub> , bla <sub>TEM-1</sub>	aac(6')-Ib, aadA1, aph(3')-XV	dfrA8, dfrB3, sul1, qnrS1, fosA, tet(C), catA1, catB2, ereA	16.0
ST517	In1444	1		bla <sub>VIM-20</sub> , bla <sub>CTX-M-3</sub> , bla <sub>OXA-1</sub> , bla <sub>TEM-1</sub>	aac(6')-Ib, armA	dfrA14, sul1, sul2, qnrB1, qnrE1, fosA, tet(A), mphE, msrE	15.0
ST533	In916	1		<i>bla</i> <sub>VIM-1</sub> , <i>bla</i> <sub>SHV-12</sub>	aac(6')-Ib, aadA1, aph(3'')-Ib, aph(6)-Id, aph(3')-XV	dfrA14, sul1, sul2, qnrS1, fosA, catB2, oqxB	14.0
ST953	In238a	1		<i>bla</i> <sub>VIM-4</sub> , <i>bla</i> <sub>CTX-M-258</sub> , <sup>b</sup> <i>bla</i> <sub>OXA-1</sub>	aac(3)-IId, aac(6')-Ib, aac(6')-Ib-cr, aadA1, aph(3'')-Ib, aph(6)-Id	dfrA14, sul1, sul2, qnrB1, fosA, tet(A)	15.0
ST1755	In916	1		bla <sub>VIM-1</sub> , bla <sub>SHV-12</sub>	aac(6')-Ib, aadA1, aph(3'')-Ib, aph(6)-Id, aph(3')-XV	sul1, sul2, qnrS1, fosA, catB2	12.0
ST1758	In916	6		bla <sub>VIM-1</sub> , bla <sub>CTX-M-15</sub> , bla <sub>SHV-12</sub> , (bla <sub>OXA-</sub> 1), (bla <sub>TEM-1</sub> )	(aac(3)-IIe), aac(6')-Ib, aadA1, (aph(3'')-Ib), (aph(6)-Id), aph(3')-XV	dfrA14, sul1, (sul2), (qnrB1), (qnrS1), (fosA), (tet(A)), catB2, (oqxA), (oqxB)	17.0
E. hormae	<i>chei</i> subsp. >	kiangfangel	nsis				
ST66	In916	8	outbreak VII	bla <sub>VIM-1</sub> , (bla <sub>CTX-M-15</sub> ), (bla <sub>SHV-12</sub> ), bla <sub>OXA-1</sub> , (bla <sub>TEM-1</sub> )	(aac(3)-lle), aac(6')-lb, aadA1, (aph(3")-lb), (aph(6)-ld), aph(3')-XV	dfrA14, sul1, sul2, (qnrB1), (qnrS1), (fosA), (catA2), catB2, (oqxB)	16.9
ST66	In916	10	outbreak VIII	bla <sub>VIM-1</sub> , (bla <sub>CTX-M-15</sub> ), (bla <sub>OXA-1</sub> ), (bla <sub>TEM-1</sub> )	(aac(3)-lle), aac(6')-lb, aadA1, (aac(6')-lb- cr), (aph(3'')-lb), aph(3')-XV	(dfrA14), sul1, (sul2), (qnrB1), (qnrS1), fosA, (tet(A)), catB2, (oqxA), (oqxB)	16.3
ST1754	In916	1		bla <sub>vIM-1</sub> , bla <sub>SHV-12</sub> , <b>bla<sub>FOX-20</sub></b> <sup>b</sup>	aac(6')-Ib, aadA1, aph(3'')-Ib, aph(6)-Id, aph(3')-XV	dfrA14, sul1, sul2, qnrB19, qnrS1, fosA, catB2, oqxB	16.0
ST92	In238	2		$bla_{VIM-4}$ , $bla_{CTX-M-15}$ , $bla_{OXA-1}$ , $bla_{TEM-1}$	aac(3)-IIe, aac(6')-Ib, aph(3'')-Ib, aph(6)-Id	dfrA14, sul1, sul2, qnrB1, fosA, tet(A), tet(D), (oqxB), (mcr-9.1)	17.0

					acquired AMR genes <sup>a</sup>		n AMR
toyo/OT	intogram	n	no po e el ce				genes/
taxa/51 ST114	Integron In238	2	remarks	blarry (blastry) (blastry)	(aac(3)-lle) aac(6')-lb (aad41) (aad42)	(dfrA12) (dfrA14) sul1 (sul2) (aprB1)	14 0
01114	11230	2		$(b a_{CTX-M-3}), (b a_{CTX-M-15}), (b a_{CTX-M-15}), (b a_{CTX-M-1})$	(aau(3))-iie), $aau(3)$ -iib, $(aauA i)$ , $(aauA 2)$ , (aph(3'')-ib) $(aph(6)$ -id) $(armA)$	fosA (tet(A)) (mphE) (msrE) (oaxB)	14.0
	In916	1		bla <sub>vim-1</sub> , bla <sub>CTX-M-15</sub> , bla <sub>OXA-1</sub> , bla <sub>TEM-1</sub>	aac(3)-Ile, aac(6')-Ib, aadA1, aph(3')-XV	dfrA14, sul1, sul2, gnrB1, gnrS1, fosA,	16.0
						tet(A), catB2	
ST121	In916	24	outbreak	bla <sub>VIM-1</sub> , bla <sub>CTX-M-15</sub> , (bla <sub>SHV-12</sub> ), (bla <sub>OXA-</sub>	(aac(3)-la), (aac(3)-lle), aac(6')-lb, aadA1,	(sat2), (dfrA1), (dfrA14), (sul1), (sul2),	18.9
			V	1), ( <i>Ыа</i> <sub>ТЕМ-1</sub> )	aph(3')-la, (aph(3'')-lb), (aph(6)-ld), aph(3')- XV	(qnrB1), (qnrS1), (fosA), (tet(A)), (catA1), catB2, (oqxA), (oqxB), (mphA)	
ST1756	In916	3		bla <sub>VIM-1</sub> , bla <sub>CTX-M-15</sub> , bla <sub>SHV-12</sub> , (bla <sub>OXA-</sub>	aac(6')-lb, aadA1, aph(3')-la, aph(3'')-lb,	sat2, dfrA1, (dfrA14), (sul1), sul2,	20.0
				<sub>1</sub> ), ( <i>bla</i> <sub>TEM-1</sub> )	aph(6)-Id, aph(3')-XV	(qnrB1), fosA, tet(A), catB2, (oqxA),	
CT101	102200	6	outbrook	hia hia hia hia	222(2) lid $222(6)$ lb $(222(6))$ lb $ar)$	(OQXB)	20 F
51121	In238a	0		DIa <sub>VIM-4</sub> , DIa <sub>CTX-M-15</sub> , DIa <sub>OXA-1</sub> , DIa <sub>TEM-1</sub>	aac(3)-110, aac(6)-10, (aac(6)-10-cr), (22dA1), 22dA2, 2pb(2'), 12, 2pb(2''), 16	Sat2, atrAI, (atrAI2), atrAI4, sui1, sui2, arrB1 fos ( tot(A) (cot(A1)) (cor(B))	20.5
			VI		(aduAT), aduA2, apri(3)-ia, apri(3)-ib, anb/6)-ld	(mnhA)	
	In238	3		blavim-4, blactx-m-15, blacxa-1, blatem-1	(aac(3)-lle), aac(6')-lb. (aac(6')-lb-cr).	sat2, dfrA1, dfrA14, sul1. (sul2). anrB1.	20.3
		-			aph(3')-la, aph(3'')-lb, aph(6)-ld, (armA)	fosA, (tet(A)), (mphE), (msrE), (oqxA),	
						oqxB	
	In2016-like	1		<i>bla</i> <sub>VIM-4</sub> , <i>bla</i> <sub>CTX-M-15</sub> , <i>bla</i> <sub>OXA-1</sub> , <i>bla</i> <sub>OXA-10</sub> ,	aac(3)-IId, aph(3')-Ia, aph(3')-VIb	sat2, arr-3, dfrA1, dfrA14, sul1, sul2,	17.0
		•		bla <sub>TEM-1</sub>		fosA, tet(A), oqxA	40.0
	In2242	2		$Da_{VIM-2}$ , ( $Da_{CTX-M-15}$ ), ( $Da_{OXA-1}$ ),	aac(3)-I, (aac(6)-Ib-cr), (aadA2), aadA6-10,	(sat2), (dtrA1), dtrA14, sul1, (sul2), tosA, tot(A) (mark E) (mark E) (sat2)	16.0
T407	In016	2		(DIA <sub>TEM-1</sub> )	(apn(3)-ia), (apn(3)-ib), apn(0)-id, (armA)	dfr (A), (mpnE), (msrE), (oqxA), (oqxB)	19.0
51407	11910	2		blavim-1, blactx-m-15, blashv-12, blaoxa-1,	aac(0)-10, aduA 1, apri(3)-7 V	tet(Δ) catB2 ogxB	10.0
	In238	2			(aac(3)-l), aac(3)-lle, aac(6')-lb, aac(6')-lb-	(sat2), (dfrA1), (dfrA12), dfrA14, sul1.	22.0
					cr, (aadA1), (aadA2), (aadA6), (aph(3')-la),	sul2, qnrB1, fosA, tet(A), (mphE), (msrE),	
					aph(3")-Ib, aph(6)-Id, (armA)	(oqxB)	
ST527	In611-like	1		bla <sub>VIM-1</sub> , bla <sub>GES-7</sub> , bla <sub>SHV-12</sub> , bla <sub>LAP-2</sub>	aac(3)-la, aac(6')-lb, aadA1,aph(6)-ld,	dfrA14, dfrB3, sul1, qnrS1, fosA, tet(A),	17.0
		•			aph(3')-XV	catB2, oqxB	40 7
ST1757	In916	3		bla <sub>VIM-1</sub> , bla <sub>CTX-M-15</sub> , (bla <sub>SHV-12</sub> ), bla <sub>OXA-1</sub>	aac(3)-IIe, aac(6')-Ib, aadA1, aph(3'')-Ib,	(sat2), (dtrA1), dtrA14, sul1, sul2,	19.7
					apn(6)-id, apn(3)-XV	(qnrBT), qnrST, tosA, tet(A), catB2, (ogxA)	
= hormae	echei subso h	offmannii				(0424)	
ST78	In2241	2		<i>bla</i> <sub>IMP-19</sub> , <i>bla</i> <sub>тем-1</sub>	(aac(3)-II), aac(6')-Ib. aadA1b. (aadA2).	dfrA15, (dfrA19), (dfrA25), sul1, fosA.	15.5
				init (c) TENET	aph(3')-la, aph(3")-lb, aph(6)-ld	catB2, oqxB, (arr)	
	In110	1		<i>bla</i> <sub>VIM-1</sub> , <i>bla</i> <sub>CTX-M-257</sub> , <sup><i>b</i></sup> <i>bla</i> <sub>LAP-2</sub> , <i>bla</i> <sub>TEM-1</sub>	aac(3)-Íld, aac(6')-Íb, aadA1, armA	dfrA15, sul1, qnrS1, fosÀ, mphE, msrE, oaxB	15.0
	In238	1		bla <sub>∨IM-4</sub>	aac(6')-lb, aac(6')-lb-cr	dfrA25, sul1, qnrB2, fosA, oqxB	8.0
ST97	In916	1		bla <sub>VIM-1</sub> , bla <sub>SHV-12</sub>	aac(6')-Ib, aadA1, aadA16, aph(3'')-Ib,	dfrA14, dfrA27, sul1, sul2, qnrB6, qnrS1,	18.0
					aph(6)-Id, aph(3')-XV	fosA, catB2, oqxB, arr-3	
ST102	In916	1		bla <sub>VIM-1</sub> , bla <sub>CTX-M-9</sub> , bla <sub>SHV-12</sub>	aac(6')-lb, aadA1, aadA2, aph(3")-lb,	dfrA16, sul1, sul2, qnrA1, fosA, tet(A),	17.0
	1-000	A			aph(6)-Id, aph(3')-XV	catB2, oqxB	47.0
	in238	1		DIa <sub>VIM-4</sub> , DIa <sub>CTX-M-15</sub> , DIa <sub>OXA-1</sub> , DIa <sub>TEM-1</sub>	aac(3)-IIE, aac(6)-ID, aac(6)-ID-cr, aadA1,	arrA14, sui1, sui2, qnrB1, tosA, tet(A),	17.0
ST104	InQ16	7		hlame hlames (hlames) (hlames)	aprilo j-ib, apriloj-iu aacl6')-lb (aacl6')-lb-cr) aadd1 (anbl3'')-	UYXD dfrA14 sul1 (sul2) (anrA1) (anrS1)	17.6
51104	111310	I		να <sub>VIM-1</sub> , να <sub>SHV-12</sub> , (να <sub>OXA-1</sub> ), (να <sub>TEM-1</sub> )	Ib), (aph(6)-Id), aph(3')-XV	fosA, (tet(D)), (catA2), catB2, (catB3),	17.0
07440	1=010	4		bla bla bla ti-		oqxB, $(arr-3)$	10.0
51118	10916	1		DIa <sub>VIM-1</sub> , DIa <sub>CTX-M-15</sub> , DIa <sub>SHV-12</sub> , DIa <sub>OXA-1</sub> ,	aac(3)-11e, aac(6)-1b, aadA1, aph(3)-XV	arra14, sul1, sul2, $qnrB1$ , $qnrS1$ , $fosA$ , $tet(\Lambda)$ , $cotP2$ , $covP$	18.0
				DIa <sub>TEM-1</sub>		iei(A), caiBZ, oqxB	

					acquired AMR genes <sup>a</sup>		n AMR
taxa/ST	integron	n isolates	remarks	resistance to β-lactams	resistance to aminoglycosides	resistance to other groups	genes/ isolate
ST135	In238	1		bla <sub>VIM-4</sub>	aac(6')-lb, aph(3')-la	sul1, fosA, ogxB	6.0
ST145	In238	1		bla <sub>VIM-4</sub> , bla <sub>SHV-12</sub> , bla <sub>OXA-10</sub>	aac(6')-lb, aadA1, aadA2, ant(2")-la	dfrA19, sul1, sul2, qnrA1, fosA, catA2, catB3, cmlA1, ogxB, mcr-9.1	17.0
ST173	In916	2		bla <sub>VIM-1</sub> , (bla <sub>CTX-M-3</sub> ), (bla <sub>CTX-M-14</sub> ), bla <sub>SHV-12</sub> , (bla <sub>TEM-1</sub> )	(aac(3)-II), (aac(6')-IIc), aac(6')-Ib, aadA1, (aph(3'')-Ib), (aph(6)-Id), aph(3')-XV	(dfrA14), (dfrA19), sul1, sul2, (qnrA1), qnrS1, fosA, catB2, oqxB, (mcr-9.1), (arr)	18.0
	In238	1		blavim-4. blashv-5. blai AP-2	aac(6')-Im. aac(6')-Ib. aph(2")-IIa.	sul1. anrS1. fosA. tet(B). catA1. oaxB	12.0
	In238a	1		bla <sub>VIM-4</sub> , bla <sub>SHV-12</sub> , bla <sub>OXA-10</sub>	aac(6')-lb, aadA1, aadA2, ant(2")-la	dfrA19, sul1, sul2, qnrA1, fosA, catA2, catB3, cmlA1, oqxB, mcr-9.1	17.0
ST316	In238	3		bla <sub>VIM-4</sub> , bla <sub>CTX-M-15</sub> , bla <sub>OXA-1</sub> , bla <sub>TEM-1</sub>	aac(3)-Ile, aac(6')-Ib, aac(6')-Ib-cr, aadA1, aph(3'')-Ib, aph(6)-Id	dfrA14, sul1, sul2, qnrB1, fosA, tet(A), oqxB	18.0
	In916	1		bla <sub>VIM-1</sub> , bla <sub>CTX-M-15</sub>	aac(6')-lb, aadA1, aph(3')-XV	sul1, fosA, catB2, oqxB	9.0
ST381	In916	1		blavim1. blashv-12	aac(6')-lb. aadA1. aph(3')-XV	dfrA14. sul1. fosA. catB2. oaxB	10.0
ST485	In238	2		<i>bla</i> <sub>VIM-4</sub> , ( <i>bla</i> <sub>SHV-12</sub> ), ( <i>bla</i> <sub>OXA-10</sub> )	aac(6')-lb, (aadA1), (aadA2), (ant(2")-la), (aph(3")-lb), (aph(6)-ld), (armA)	(dfrA19), sul1, sul2, (qnrA1), fosA, catB3, (cmlA1), (mphE), (msrE), oaxB, mcr-9.1	15.5
ST764	In916	1		bla <sub>VIM-1</sub> , bla <sub>CTX-M-15</sub> , bla <sub>OXA-1</sub> , bla <sub>TEM-1</sub>	aac(3)-Ile, aac(6')-Ib, aadA1, aph(3')-XV	dfrA14, sul1, sul2, qnrB1, qnrS1, fosA, tet(A), catB2, ogxB	17.0
ST1641	In916	1		bla <sub>VIM-1</sub> , bla <sub>SHV-12</sub>	aac(6')-lb, aadA1, aph(3'')-lb, aph(6)-ld, aph(3')-XV	dfrA14, sul1, sul2, qnrS1, fosA, catB2, oaxB	14.0
ST1753	In916	3		bla <sub>VIM-1</sub> , bla <sub>SHV-12</sub>	aac(6')-lb, aadA1, aph(3")-lb, aph(6)-ld, aph(3')-XV	dfrA14, sul1, sul2, qnrS1, fosA, catB2, oaxB	15.0
ST1759	In916	2		bla <sub>VIM-1</sub> , bla <sub>CTX-M-15</sub> , bla <sub>OXA-1</sub> , bla <sub>TEM-1</sub>	aac(3)-Ile, aac(6')-Ib, aadA1, aph(3')-XV	dfrA14, sul1, sul2, (qnrB1), qnrS1, fosA, tet(A), catB2, oqxB	17.5
E. hormae	<i>chei</i> subsp. d	oharae					
ST68	In238	1		bla <sub>VIM-4</sub> , bla <sub>CTX-M-3</sub> , bla <sub>TEM-1</sub>	aac(3)-IId, aac(6')-Ib, armA	sul1, mphE, msrE, oqxB	10.0
ST94	In70	1		<i>bla</i> <sub>VIM-1</sub> , <i>bla</i> <sub>SHV-12</sub> , <i>bla</i> <sub>OXA-10</sub>	aac(6')-lb, aadA1, aadA2, ant(2'')-la, aph(3')-XV	sat2, dfrA1, dfrA14, dfrA19, sul1, sul2, qnrS1, catA2, catB2, catB3, cmlA1, mcr- 9.1	20.0
	In238	1		bla <sub>∨IM-4</sub>	aac(6')-lb, aadA1, aph(3'')-lb, aph(6)-ld, armA	sat2, dfrA1, sul1, qnrS1, tet(A), mphE, msrE, oqxA	14.0
ST108	In1008	8		<i>bla</i> <sub>VIM-2</sub> , ( <i>bla</i> <sub>CTX-M-9</sub> )	aac(6')-Ib, (aadA2)	(dfrA16), sul1, tet(A), oqxB	6.7
E. hormae	<i>chei</i> subsp. <i>I</i>	hormaeche	i				
ST528 E. roggenk	In238 In238	1		bla <sub>VIM-4</sub> , bla <sub>CTX-M-15</sub> , bla <sub>OXA-1</sub>	aac(3)-lle, aac(6')-lb	dfrA14, qnrB1, tet(A)	8.0
ST95	In238	1		bla <sub>viM-4</sub>	aac(6')-Ib	sul1, fosA, tet(A), oqxB	6.0
ST96	In238	4		bla <sub>VIM-4</sub> , (bla <sub>CTX-M-3</sub> ), (bla <sub>GES-7</sub> ), (bla <sub>SHV-12</sub> ), (bla <sub>OXA-1</sub> ), (bla <sub>OXA-10</sub> ), (bla <sub>TEM-1</sub> )	(aac(3)-IId), aac(6')-Ib, (aadA2), (ant(2'')- Ia), (aph(3')-Ia), (aph(3')-Ib), (aph(6)-Id), (armA)	(dfrA12), (dfrA19), (dfrB3), sul1, (qnrS2), fosA, (catA2), (catB3), (mphE), (msrE), (oqxA), (oqxB), (mcr-9,1), (arr-3)	14.5
ST166	In916	1		bla <sub>VIM-1</sub>	aac(6')-lb, aadA1, aph(3'')-lb, aph(6)-ld, aph(3')-XV	sul1, sul2, qnrS1, fosA, catB2, oqxB	12.0
ST523	In238	1		bla <sub>∨IM-4</sub>	aac(6')-Ib. armA	sul1, fosA, mphE. msrE	7.0
ST1761 E. asburiae	In238	1		bla <sub>VIM-4</sub>	aac(6')-lb	sul1, fosA, oqxB	5.0
ST23	In238	1		blayima	aac(6')-Ib	sul1. fosA	4.0
ST25	In238a	1		bla <sub>VIM-4</sub> , bla <sub>CTX-M-3</sub> , bla <sub>OXA-1</sub> , bla <sub>TEM-1</sub>	aac(6')-lb, aadA1, aadA2, aph(3')-Vla	dfrA14, qnrB1, qnrS1, fosA, tet(A), tet(C), oqxB	15.0

E. kobei

					acquired AMR genes <sup>a</sup>		n AMR
		n					genes/
taxa/ST	integron	isolates	remarks	resistance to β-lactams	resistance to aminoglycosides	resistance to other groups	isolate
ST99	In238	1		bla <sub>VIM-4</sub> , bla <sub>SHV-5</sub>	aac(6')-Im, aac(6')-Ib, aadA1, aph(2")-IIa, aph(3')-VI	dfrA1, sul1, fosA, tet(B), catA1, oqxB	13.0
<i>E. ludwigii</i> ST1306	In916	1		bla <sub>VIM-1</sub> , bla <sub>SHV-12</sub>	aac(6')-lb, aadA1, aph(3'')-lb, aph(6)-ld, aph(3')-XV	dfrA14, sul1, sul2, qnrS1, fosA, catB2, oqxB	14.0
E. mori ST1760	In238a	1		bla <sub>vim-4</sub> , bla <sub>ctx-M-9</sub>	aac(6')-lb, aac(6')-lb-cr, aadA2, ant(2'')-la	dfrA16, sul1, qnrA1, qnrE1, fosA, oqxB	12.0

a – symbols in parentheses refer to the genes that occurred not in all isolates of the corresponding genotypes.
 b – new blactx.m and blaFoX genes are indicated in bold; nucleotide sequences of new genes are available under the following GenBank accession numbers: blactx.m.256, OP081688; blactx.m.258, OP346113; blaFoX.20, OP297845; blactx.m.257, OP297846.

			AMR genes located on short	
Isolate			contigs not assigned to individual	
(ST)	Plasmids with <i>bla</i> VIM gene	Other plasmids	plasmids or chromosome	Chromosome
743/14	IncA: bla <sub>VIM-1</sub> , bla <sub>SHV-12</sub> , aac(6')-lb, aadA1,	IncHI2+HI2A: bla <sub>OXA-1</sub> , aac(6')-lb-cr,	-	bla <sub>CTX-M-15</sub> , bla <sub>TEM-1</sub> (x2), aph(3')-la,
(ST121)	aph(3")-Ib, aph(3')-XV, aph(6)-Id, mphA, catB2, sul1, sul2, qnrS1, dfrA14	aadA1, catA1, qnrB1, tet(A), dfrA14		aph(6)-Id, aph(3")-Ib, sul2, dfrA1, fosA, oqxB, sat2
5955/16	IncA: bla <sub>VIM-1</sub> , aac(6')-lb, aadA1, aph(3")-lb,	IncFII+FIB: bla <sub>OXA-1</sub> , aac(6')-lb-cr, catA2,	-	fosA, oqxB
(ST66)	aph(3')-XV, aph(6)-Id, catB2, sul1, sul2, qnrS1,	qnrB1, dfrA14		
7753/18	IncA: bla <sub>VIM-1</sub> , bla <sub>SHV-12</sub> , aac(6')-lb, aph(3")-lb,	IncFII+FIB: bla <sub>OXA-1</sub> , aac(6')-lb-cr,	bla <sub>CTX-M-15</sub> , bla <sub>TEM-1</sub> , aac(3)-lle,	aadA1, dfrA1, fosA
(ST89)	aadA1, aph(3')-XV, aph(6)-Id, mphA, catB2, sul1, sul2, dfrA14	aadA1, catB3, catA1, tet(A)	aph(6)-Id, aph(3")-Ib, sul2	
4969/09	IncHI2+HI2A: blavim-4, aac(6')-lb, catA2, sul1,	-	bla <sub>OXA-10</sub> , aac(6')-lb, aadA1,	bla <sub>CTX-M-3</sub> (x3), fosA
(ST90)	mcr-9.1		aadA2, ant(2")-la, catB3, sul1	
0004/00			(x2), dfrA19, cmIA1	
6234/09	IncFII+FIA: bla <sub>VIM-4</sub> , aac(6')-lb	-	-	$bla_{CTX-M-3}(2x), bla_{TEM-1}, aac(3)-IIa,$
(5190)				apn(3")-ib, aadA1, dfrA1, sul1 (2x), fosA
5435/13	IncN3: bla <sub>VIM-4</sub> , aac(6')-lb, sul1	IncHI2+HI2A: bla <sub>CTX-M-15</sub> , bla <sub>OXA-1</sub> ,	-	fosA, oqxB
(ST134)		aac(6')-lb-cr, aac(3)-lle, aadA1, catA1, qnrB1, tet(A), dfrA14		
5713/17	IncFIB:bla <sub>VIM-4</sub> , bla <sub>TEM-1</sub> , aac(3)-lld, aac(6')-lb,	IncHI2+HI2A: bla <sub>OXA-1</sub> , aac(6')-lb-cr,	-	bla <sub>CTX-M-15</sub> , bla <sub>TEM-1</sub> (x2), aph(3')-la,
(ST121)	aadA2, mphA, sul1 (x2), dfrA12	aadA1, catA1, qnrB1, tet(A), dfrA14		aph(6)-Id, aph(3")-Ib, sul2, dfrA1, fosA,
				oqxB, sat2
8770/11 (ST89)	-	-	-	bla <sub>CTX-M-3</sub> , bla <sub>VIM-40</sub> , aac(6')-lb, aph(3'')-lb, aph(3')-VI, aph(6)-ld, sul1, fosA
2944/06	-	-	-	bla <sub>CTX-M-3</sub> (x2),bla <sub>VIM-20</sub> , aac(3)-IId,
(ST89)				aac(6')-lb, aph(3')-la, aadA2, armA (x2),
				sul1(x2), dfrA12, fosA, msr(E) (x2),
				mph(A), mph(E) (x2)

#### Appendix Table 10. Resistomes of the isolates subjected to the long-read WGS analysis

Appendix Table 11. Repli	con types of the	Enterobacter spp.	isolates
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taxa/ST	integron	n isolates	replicon types <sup>a</sup>	n replicon types/ isolate
E. hormaeche	i subsp. steigerwaltii			
ST45	In238	7	(A), FIB, FII, HI2, HI2A, CoIRNAI	5.3
	In916	2	A, FÌB, (M1), Col440I, Col440II, ColRNAI	5.0
ST62	In916	1	A, Ń, Col440II, ColRNAI	4.0
ST89	In916	48	A, (FIA), (FII), (HI1A), (HI1B), (HI2), (HI2A), (M1), (Col440II), ColRNAI	4.5
	In238	3	(A), (HI2), (HI2A), (M1), ColRNAI	1.7
	In1654	1	FIÌ, HIŻ, HIŻA, M2, Col440II, ColRNAI	6.0
	In2238	1	HI2, HI2A	2.0
	In1445	7	(FII)	0.1
	In1008	2	M1. ColRNAL	2.0
	In1444	12	(FII), ColpVC	1.1
ST90	In238	107	(A), (FIA), (FIB), (FII), (HI1A), (HI1B), (HI2), (HI2A), (M1), (M2), (N), (U), (Col440II), (ColRNAI), (ColpVC), (ColpWES), (p0111)	4.1
	In238a	5	FIA, FII, (HI2), (HI2A), (Col440)), (Col440II), (ColRNAI)	3.6
	In238a: In2241	2	HI2, HI2A, N3, (Col44011), ColRNAI	4.5
	In916	1	A, HI2, HI2A, Col440II, ColRNAI	5.0
	In2240	1	A. FIB. Col440I. Col440II	4.0
	In237a	1	FIB. M2. CoIRNAI	3.0
ST91	In1008	2	HI2. HI2A. M2	3.0
	In70	3	A. (N). (HI2). (Col440II). (ColRNAI). (ColpVC)	3.7
	In916	1	Α	1.0
ST93	In238	2	FIB. (FII). (HI2). (HI2A). (R). Col440IL ColRNAI	5.0
	In1008	1	FII. M2. R. Col4401. ColRNAI	5.0
ST106	In916	1	A. (FIB). HI2. HI2A. Col440IL ColRNAI	5.3
ST110	In916	1	A. ColRNAI	2.0
ST116	In2240	1	A	1.0
ST134	In238	9	HI2, HI2A, N3, (ColRNAI)	3.1
	In238a	2	N3	1.0
	In916	2	Α	1.0
ST175	In238a	2	HI2. HI2A. N3	3.0
	In916	1	A. HI2. HI2A	3.0
ST184	In916	1	A, HI2, HI2A, Q2	4.0
ST517	In1444	1	FIB, FII, M2, COIRNAI	4.0
ST533	In916	1	A	1.0
ST953	In238a	1	FIB, HI2, HI2A, ColpVC	4.0
ST1755	In916	1	A, FIB, FII, Col440I, Col440II	5.0
ST1758	In916	6	(A), (HI2), (HI2A), Col440I, Col440II, (ColRNAI)	5.2
ST1762	In238	2	FIA, FIB, (FII), HI2, HI2A, Col440II, ColRNAI	6.5
E. hormaeche	i subsp. xiangfangensi	s		
ST66	In916	18	A, (FIB), (FII), (HI2), (HI2A), (M1), (Col440II), ColRNAI	4.2
ST92	In238	2	FIB, FII, M1, Col440I, Col440II, ColRNAI	6.0
ST114	In238	2	(HI1A), (HI1B), (HI2), (HI2A), (M2), (N3), (Col440II), ColRNAI	4.5
	In916	1	A, FIB, HI2, HI2A, Col440II, ColRNAI	6.0
ST121	In916	24	A, FIB, (FII), (HI2), (HI2A), (M1), (R), (Col440II), (ColRNAI), (ColpVC)	5.0
	In238a	6	FIB, HI2, HI2A, CoIRNAI	4.0
	In238	3	(C), FIB, HI2, HI2A, (N3), (CoIRNAI), (ColpVC)	4.7

				n replicon
				types/
taxa/ST	integron	n isolates	replicon types <sup>a</sup>	isolate
	In2016-like	1	C, FIB, FII, HI2, HI2A, Col440II, ColRNAI, ColRGK	8.0
	In2242	2	FIB, FII, HI2, HI2A, (M1), (Col440II), ColRNAI	6.0
ST407	In916	2	A, HI2, HI2A, ColRNAI	4.0
	In238	2	(C), HI2, HI2A, (N3), Col440II, ColRNAI	5.0
ST527	In611-like	1	A, M1, Q2, ColRNAI	4.0
ST1754	In916	1	A. Q2. Col440	3.0
ST1756	In916	3	A. FIB. HI2. HI2A. Col44011. ColRNAI	6.0
ST1757	In916	3	A FU HIZ HIZA (M1) (B) (Col4401) ColBNAL	6.7
E hormaeche	i subsp hoffmannii	0	, , , , , , , , , , , , , , , , , , ,	0.1
ST78	In2241	2	FIB (FII) HI2 HI2A Col4401 Col4401	5.5
0110	In110	1		3.0
	In 238	1		3.0
ST07	In230	1		5:0 4 0
ST37 ST102	In910	1		4.0
31102	11910	1		5.0
07404	11230	1		0.0
ST104	10916	1	A, (FIB), (FIB <sub>K</sub> ), (FII), (HITA), (HITB), (N), (N2), (Col44011), ColKINAI	5.3
51118	10916			4.0
ST135	In238	1	Col4401, Col4401, ColRNA	3.0
ST145	In238	1	FIB, FII, HI2, HI2A, N3, CORNAI	6.0
ST173	In916	2	A, (HI2), (HI2A), (M2), (N), Col440II, ColRNAI	5.0
	In238	1	A, Col440I, Col440II, ColRNAI	4.0
	In238a	1	HI2, HI2A, N, N3	4.0
ST316	In238	3	(FIB), HI2, HI2A, N3, R, COIRNAI	5.3
	In916	1	A, R, ColRNAI	3.0
ST381	In916	1	A, FIB, FII, R, Col440I, ColRNAI	6.0
ST485	In238	2	(C), FIB, (FII), HI2, (HI2A), (N3), (R), (Col440II), ColRNAI	6.0
ST764	In916	1	A, FIB, HI2, HI2A, CoIRNAI	5.0
ST1641	In916	1	A, FIB, ColRNAI	3.0
ST1753	In916	3	A, ColRNAI	2.0
ST1759	In916	2	A, HI2, HI2A, CoIRNAI	4.0
E. hormaeche	i subsp. <i>oharae</i>			
ST68	In238	1	ColRNAI	1.0
ST94	In70	1	A. HI2. HI2A	3.0
	In238	1	C. N. ColRNAI	3.0
ST108	In1008	8	FIB (FII) (HI2) (HI2A) M1 R Col440II ColRNA	64
ST528	In238	1	HIA HIA MI COBNAL	4.0
ST95	In238	1	FIB FIL M1 COLARD COLENAL DENTASO2	6.0
ST96	In238	4	(H(2), (H(2A), (M,2), (O2), (B), (U), Collect)	3.2
ST166	In230	1	$A = EB_{c}$ + 11A + 11B Col44011 ColENAL pENTAS02	3. <u>2</u> 7 0
ST523	In238	1		1.0
ST1761	In238	1		5.0
5 ashuriae	11250	I		5:0
L. asuunat	10000	1		4.0
0120	111230	1		4.0
SIZJ E kohoi	111238a	I	ГІД, ГІІ, ПІZ, ПІZA, IN, GUI44UII, GUIKINAI, PEINTASUZ	8.0
	1=000	4		6.0
5199	IN238	1	A, FIB <sub>K</sub> , FII, COI44UII, COIKINAI, $pKPC$ -CAV1321	6.0
∟. iuawigii				

				n replicon		
				types/		
taxa/ST	integron	n isolates	replicon types <sup>a</sup>	isolate		
ST1306	In916	1	A	1.0		
E. mori						
ST1760	In238a	1	FII, HI2, HI2A, N3, Col440I, Col440II, ColRNAI	7.0		
a compared in parentheres refer to the replicent times that accurred not in all isolates of the companying genetimes						

<sup>a</sup> – symbols in parentheses refer to the replicon types that occurred not in all isolates of the corresponding genotypes.



**Appendix Figure 1.** VIM/IMP-producing Enterobacterales in Poland; A) regional, and B) annual distribution



**Appendix Figure 2.** Geographic distribution and clonal analysis of *E. hormaechei* ST89 in Poland. A) Geographic distribution of the isolates shown on the map of the country with main administrative regions. Circles represent medical centres where the isolates were recorded. Sizes of the circles are proportional to numbers of cases. B) SNP-based minimum spanning tree of the isolates. Lengths of branches are related to numbers of SNPs between linked isolates. Numbers of SNPs are indicated above the branches or next to the dots.



**Appendix Figure 3.** Geographic distribution and clonal analysis of *E. hormaechei* CC121 (ST121 and ST1756) in Poland. A) Geographic distribution of the isolates shown on the map of the country with main administrative regions. Circles represent medical centres where the isolates were recorded. Sizes of the circles are proportional to numbers of cases. B) SNP-based minimum spanning tree of the isolates. Lengths of branches are related to numbers of SNPs between linked isolates. Numbers of SNPs are indicated above the branches or next to the dots.



**Appendix Figure 4.** Geographic distribution and clonal analysis of *E. hormaechei* CG66 (ST66 and ST1754) in Poland. A) Geographic distribution of the isolates shown on the map of the country with main administrative regions. Circles represent medical centres where the isolates were recorded. Sizes of the circles are proportional to numbers of cases. B) SNP-based minimum spanning tree of the isolates. Lengths of branches are related to numbers of SNPs between linked isolates. Numbers of SNPs are indicated above the branches or next to the dots.



**Appendix Figure 5.** Geographic distribution and clonal analysis of *E. hormaechei* ST134 in Poland. A) Geographic distribution of the isolates shown on the map of the country with main administrative regions. Circles represent medical centres where the isolates were recorded. Sizes of the circles are proportional to numbers of cases. B) SNP-based minimum spanning tree of the isolates. Lengths of branches are related to numbers of SNPs between linked isolates. Numbers of SNPs are indicated above the branches or next to the dots.



**Appendix Figure 6.** SNP-based phylogenetic tree of all study Polish *E. hormaechei* ST90 isolates compared with the international ST90 genomes available in GenBank. Numbers in the inner circle correspond to original numbers of the study isolates or GenBank assembly numbers. The presence of carbapenemases is indicated in the outer circles using corresponding colors. The country of origin other than Poland is presented with country codes: AU, Australia; CN, China; DE, Germany; EC, Ecuador; FR, France; GB, Great Britain; JP, Japan; PT, Portugal; RO, Romania; TW, Taiwan; ZA, South Africa. The tree was constructed using Parsnp and visualized with iTOL.



**Appendix Figure 7.** SNP-based phylogenetic tree of all study Polish *E. hormaechei* ST89 isolates compared with the international ST89 genomes available in RefSeq. Numbers in the inner circle correspond to original numbers of the study isolates or RefSeq assembly numbers. The presence of carbapenemases is indicated in the middle circle using corresponding colors. The region of Poland where isolates were recorded is presented in outer circle. DE, Germany. The tree was constructed using Parsnp and visualized with iTOL.



**Appendix Figure 8.** SNP-based phylogenetic tree of all study Polish *E. hormaechei* ST121 isolates compared with the international ST121 genomes available in RefSeq. Numbers in the inner circle correspond to original numbers of the study isolates or RefSeq assembly numbers. The presence of carbapenemases is indicated in the outer circles using corresponding colors. The country of origin other than studied isolates is presented with country codes: BR, Brazil; DE, Germany; MA, Morocco; PK, Pakistan; PL, Poland; UG, Uganda. The tree was constructed using Parsnp and visualized with iTOL.



**Appendix Figure 9.** SNP-based phylogenetic tree of all study Polish *E. hormaechei* ST66 isolates compared with the international ST66 genomes available in RefSeq. Numbers in the inner circle correspond to original numbers of the study isolates or RefSeq assembly numbers. The presence of carbapenemases is indicated in the outer circles using corresponding colors. The country of origin other than Poland is presented with country codes: AU, Australia; CA, Canada; CN, China; CO, Colombia; DE, Germany; EG, Egypt; ES, Spain; FR, France; GB, Great Britain; NG, Nigeria; TW, Taiwan; TG, Togo; US, United States; VN, Vietnam. The tree was constructed using Parsnp and visualized with iTOL.



**Appendix Figure 10.** SNP-based phylogenetic tree of all study Polish *E. hormaechei* ST134 isolates compared with the international ST134 genomes available in RefSeq. Numbers in the inner circle correspond to original numbers of the study isolates or RefSeq assembly numbers. The presence of carbapenemases is indicated in the outer circles using corresponding colors. The country of origin other than Poland is presented with country codes: CN, China; GB, Great Britain; IR, Iran; LB, Lebanon; US, United States. The tree was constructed using Parsnp and visualized with iTOL.



**Appendix Figure 11.** Comparison of the VIM-4-encoding (integron In238) IncHI2+HI2A p4969H plasmid (inner, thin black circle) to previously reported plasmids of the highest homology: p17277A\_477 (Argentina; CP043927), pIMP-4-EC62 (China; MH829594), pIMPIncHI2\_334kb (United Kingdom; CP044215), pMOL952\_IncHI2 (Italy; OU015717), pSPRC-Echo1 (Australia; CP032842), p49790\_MCR (Czechia; CP059425), p1-Ec387 (Japan; AP024583) and P1 (Spain; OW849504). The outer thick black ring refers to the annotation of p4969H, with the selected genes indicated. The percentage of sequence identity is reflected by color intensity. The picture was created using BRIG software (http://brig.sourceforge.net/).



**Appendix Figure 12.** Comparison of the VIM-4-encoding (integron In238) IncFII+FIA p6234F plasmid (inner, thin black circle) to previously reported plasmids of the highest homology: p58983CZ\_IncFII (Czechia; CP085735), pRHBSTW-00399\_2 (United Kingdom; CP056561), plasmid P1 (Spain; OW849321), plasmid P1 (Spain; OW849504), plasmid P1 (Spain; OW968027), plasmid unnamed1 (United States; CP026851), pRHBSTW-00508\_2 (United Kingdom; CP056442), pRHBSTW-00542\_2 (United Kingdom; CP056712), pMB6956\_1 (United States; CP103612). The outer thick black ring refers to the annotation of p6234F, with the selected genes indicated. The percentage of sequence identity is reflected by color intensity. The picture was created using BRIG software (http://brig.sourceforge.net/).



**Appendix Figure 13.** Comparison of sequences of the In238-containing Tn*21*-like elements, Tn*7536*, from p6234F and p4969H, and the most similar transposon Tn*1696* with the In4 integron (U12338). The percentage of sequence identity is reflected by gray color intensity. Individual loci (antibiotic resistance, mobile genetic elements, heavy metal resistance genes) are marked by colored arrows or triangles as explained below. The picture was created using the Easyfig 2.2.5 software.



**Appendix Figure 14.** Comparison of the VIM-4-encoding (integron In238a) IncFIB p5713F plasmid (inner, thin black circle) to previously reported plasmids of the highest homology: P2 (Spain; OW849210), pOXA1\_SCLZS47 (China; CP092496), pCFR17\_1 (China; CP035277), plasmid unnamed2 (Sierra Leone; CP017059), plasmid P2 (Spain; OW969877), pC45\_002 (Australia; CP042553), pC50\_002 (Australia; CP042480) and pE33\_002 (Australia; CP042519). The outer thick black ring refers to the annotation of p5713F, with the selected genes indicated. The percentage of sequence identity is reflected by color intensity. The picture was created using BRIG software (http://brig.sourceforge.net/).



**Appendix Figure 15.** Comparison of the VIM-4-encoding (integron In238) IncN p5435N plasmid (inner, thin black circle) to previously reported plasmids of the highest homology: pBD-50-Eh-VIM-1 (Switzerland; CP063227), pUR-EC3.3 (Uruguay; MZ382872), p17511\_70 (Argentina; MN583554), pEC448\_OXA163 (Argentina; CP015078), p0801-IMP (China; KT345947), p7121-IMP (China; KX784502), p17285-IMP (China; KX784503), pY-2.p1 (China; CP090797), pC52\_003 (Australia; CP042548), pJIE137 (Australia; EF219134), plasmid:3 (Australia; LR890351), pIMPIncN3\_52kb (United Kingdom; CP043856). The outer thick black ring refers to the annotation of p5435N, with the selected genes indicated. The percentage of sequence identity is reflected by color intensity. The picture was created using BRIG software (http://brig.sourceforge.net/).



**Appendix Figure 16.** Comparison of the VIM-1-encoding (integron In916) IncA p743A plasmid (inner, thin black circle) to p7753A, p5955A, and previously reported plasmids of the highest homology: pRIVM0001\_VIM-1 (The Netherlands; MH220284), pKC-BO-N1-VIM (Italy; MG228427), pGA\_VIM (Italy; MN783743), pFDL-VIM (Italy; MN783744), p550\_IncA\_VIM\_1 (Italy; CP058224), p9546/19\_1 (Poland; ON081626), pEco15-1 (Canada; CP047711), pIncAC-KP4898 (Italy; KY882285), pR210-2-VIM (China; CP034084), pIB2020\_IncA (Italy; CP059480), pIBAC\_Incx3\_A/C (Italy; MH594478), pIncA\_P7699 (France; CP071911), pIncA\_P7536 (France; CP071789), pIBAC\_IncA/C (Italy; MH594477), pRA1 (United States; FJ705807). The outer rings refers to the annotation of p743A, with the selected genes indicated. The percentage of sequence identity is reflected by color intensity. The picture was created using BRIG software (http://brig.sourceforge.net/).



**Appendix Figure 17.** Comparison of the resistance regions of In916-carrying IncA-like plasmids characterized by MinION sequencing in the study (p743A, p7753A and p5955A) with the corresponding part of the pGA\_VIM plasmid reported in Italy (MN783743). The percentage of sequence identity is reflected by gray color intensity. Individual loci (antibiotic resistance genes, mobile genetic elements, heavy metal resistance genes and integration sites) are marked by colored arrows or triangles as explained below. The picture was created using the Easyfig 2.2.5 software.



**Appendix Figure 18.** Comparison of the *Eh*Gl3 sequence with the reference *clc*-type element (GenBank acc. No. AJ617740; Gaillard M, et al. J Bacteriol. 2006; 188:1999-2013) and the GI with the highest homology PAGI-15 (GenBank acc. No. KX196168; Hong JS, et al. Antimicrob Agents Chemother. 2016; 60:7216-23). The percentage of sequence identity is reflected by gray color intensity. Individual loci (antibiotic resistance, mobile genetic elements, heavy metal resistance genes, integrase genes) are marked by coloured arrows or triangles as explained below. The picture was created using the Easyfig 2.2.5 software.