

# Polyclonal Dissemination of OXA-232 Carbapenemase-Producing *Klebsiella pneumoniae*, France, 2013–2021

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During 2013–2021, increased prevalence of oxacillinase 232-producing Enterobacterales was observed in France, mostly driven by its emergence in *Klebsiella pneumoniae*. Whole-genome sequencing identified that oxacillinase 232-producing *K. pneumoniae* belonged to 14 sequence types (STs), among which 2 polyclonal high-risk clones, ST-231 and ST-2096, were overrepresented.

The massive dissemination of carbapenemase-producing Enterobacterales poses a global threat to public health. Carbapenem antibiotics remain the last line of defense against highly resistant Enterobacterales. Carbapenemases have been identified in 3 of the 4 classes of the Ambler classification: class A carbapenemases (mostly *Klebsiella pneumoniae* carbapenemase types) (1), class B carbapenemases or metallo- $\beta$ -lactamases (mostly New Delhi metallo- $\beta$ -lactamase [NDM], Verona integron-mediated metallo- $\beta$ -lactamase [VIM], or imipenemase types) (2), and class D carbapenemases (mostly oxacillinases [OXAs] of OXA-48 types) (3). In France, the most prevalent carbapenemases are of OXA-48 type (4). According to the Beta-Lactamase Database (<http://www.bldb.eu>), >50 OXA-48-like carbapenemase variants have

been identified. OXA-48, OXA-162, OXA-181, OXA-232, OXA-204, and OXA-244 are the most common enzymes identified among these carbapenemases (4).

OXA-232 differs from OXA-181 by a single amino acid substitution (Arg214Ser), differing itself from OXA-48 by 4 substitutions (Thr104Ala, Asn110Asp, Glu168Gln, and Ser171Ala). OXA-232 has been demonstrated to possess a weaker hydrolytic activity toward carbapenems but a stronger ability to hydrolyze penicillins compared with OXA-48 and OXA-181 (5,6). The *bla*<sub>OXA-232</sub> gene usually is located on a 6-kb nonconjugative ColE-type plasmid within a truncated Tn2013-like transposon (5). Furthermore, the genetic environment surrounding the *bla*<sub>OXA-232</sub> gene is comparable to that of the *bla*<sub>OXA-181</sub> gene, suggesting that OXA-232 is derived directly from OXA-181 (4).

Previous research has mainly identified OXA-232 in *Escherichia coli* and *K. pneumoniae* isolates and has found that this variant is endemic in China, India, South Korea, and Thailand (4,7,8). For *K. pneumoniae*, several outbreaks have been reported with different sequence types (STs), including ST-14, ST-15, ST-16, ST-23, ST-231, and ST-437 (4,9–11). Moreover, to the best of our knowledge, there are no data from France regarding OXA-232 outbreaks and epidemiology since the first description of 1 *E. coli* ST-2968 and 2 *K. pneumoniae* ST-14 isolates from patients returning to France from India in 2012 (5).

In addition, strains coproducing NDM and OXA-232 have been reported in several countries (12–14). In these strains, *bla*<sub>NDM</sub> and *bla*<sub>OXA-232</sub> are carried by 2 different plasmids (13). The *bla*<sub>OXA-232</sub> gene is located on a ColE-type plasmid, whereas the *bla*<sub>NDM</sub> gene usually is carried by an *incF*-type plasmid (8).

Given the increasing prevalence of OXA-232-producing Enterobacterales in Europe, it is crucial to better understand the driving forces of such

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dissemination. In this study, we used whole-genome sequencing to decipher the epidemiology of OXA-232–producing *K. pneumoniae* in France during 2013–2021.

### The Study

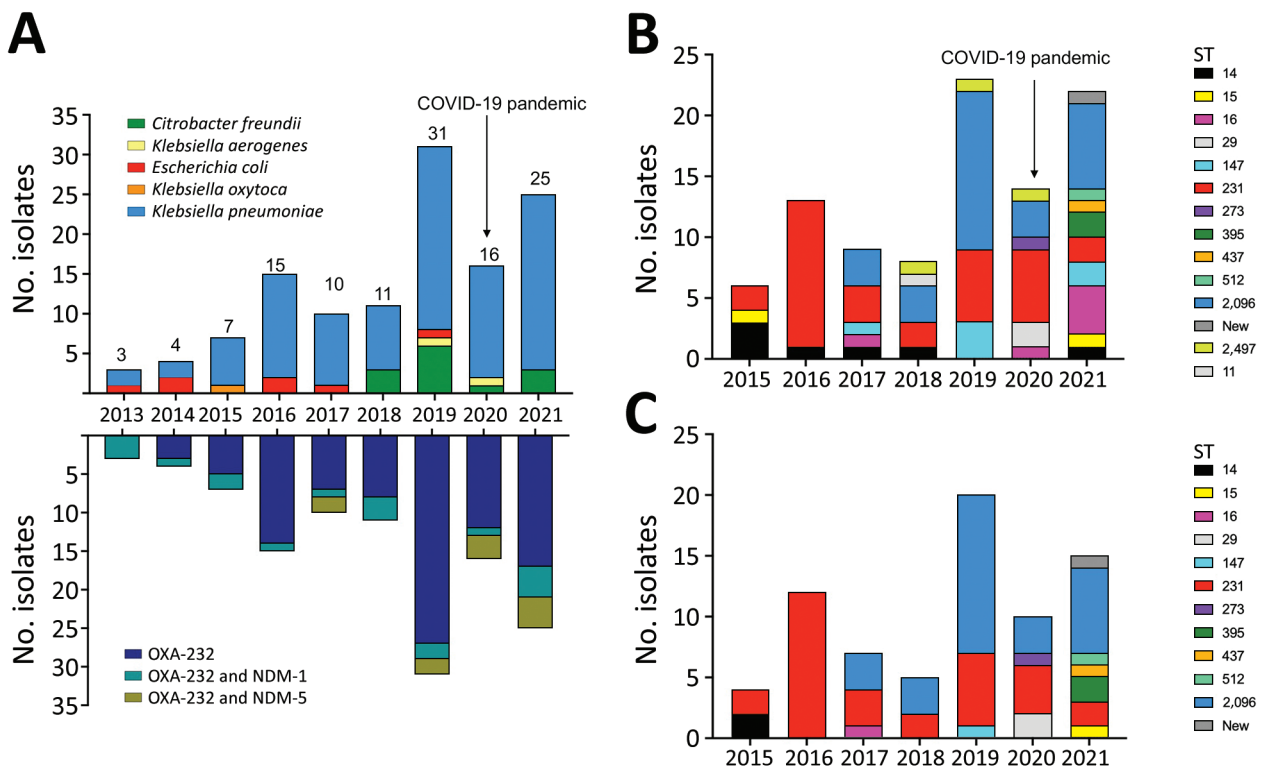
During 2013–2021, France's National Reference Centre received 122 nonduplicate OXA-232–producing Enterobacteriales, including 99 *K. pneumoniae*, 13 *Citrobacter freundii*, 7 *E. coli*, 2 *K. aerogenes*, and 1 *K. oxytoca* (Figure 1, panel A; Appendix Table 1, <https://wwwnc.cdc.gov/EID/article/28/11/20-1040-App1.pdf>). These clinical isolates were cultured from rectal swabs (n = 92), urine samples (n = 18), blood cultures (n = 2), respiratory tracts samples (n = 1), and other or unknown origins (n = 9) (Appendix Table 1).

Among these strains, 16 coproduced NDM-1 and 9 coproduced NDM-5 (Figure 1, panel A). Overall, the prevalence of OXA-232 among OXA-48–like producers was significantly higher during 2019–2021 (1.33% among OXA-48–like) compared to 2013–2018 (0.70% among OXA-48–like) ( $\chi^2$  test,  $p < 0.05$ ) (Figure 1, panel A; Table 2). The prevalence of NDM and OXA-232–coproducing isolates also slightly increased (0.15% among NDM and 0.27% among OXA-48–like from

2013–2018 to 2019–2021) (Figure 1, panel A; Appendix Table 2).

We performed short-read next-generation sequencing on all *K. pneumoniae* strains producing OXA-232 during 2015–2021 (n = 95) using a HiSeq system (Illumina, <https://www.illumina.com>) and submitted them to GenBank (Appendix Table 1). We assembled Illumina reads using shovill 1.1.0 (<https://github.com/tseemann/shovill>) and SPAdes 3.14.0 (<http://bioinf.spbau.ru/spades>) multilocus sequence typing programs, and we performed resistome analysis using pubMLST (<https://pubmlst.org>) and Resfinder (<https://cge.cbs.dtu.dk/services/ResFinder>). For phylogenetic analysis, we mapped next-generation sequencing reads to the reference genome (*K. pneumoniae* HS11286 [GenBank accession no. NC\_016845.1]) using SNIppy 4.6.0 (<https://software.cqls.oregonstate.edu/updates/snippy-4.6.0>). We visualized metadata and phylogenetic trees using iTOL 6.5.2 (<https://itol.embl.de>).

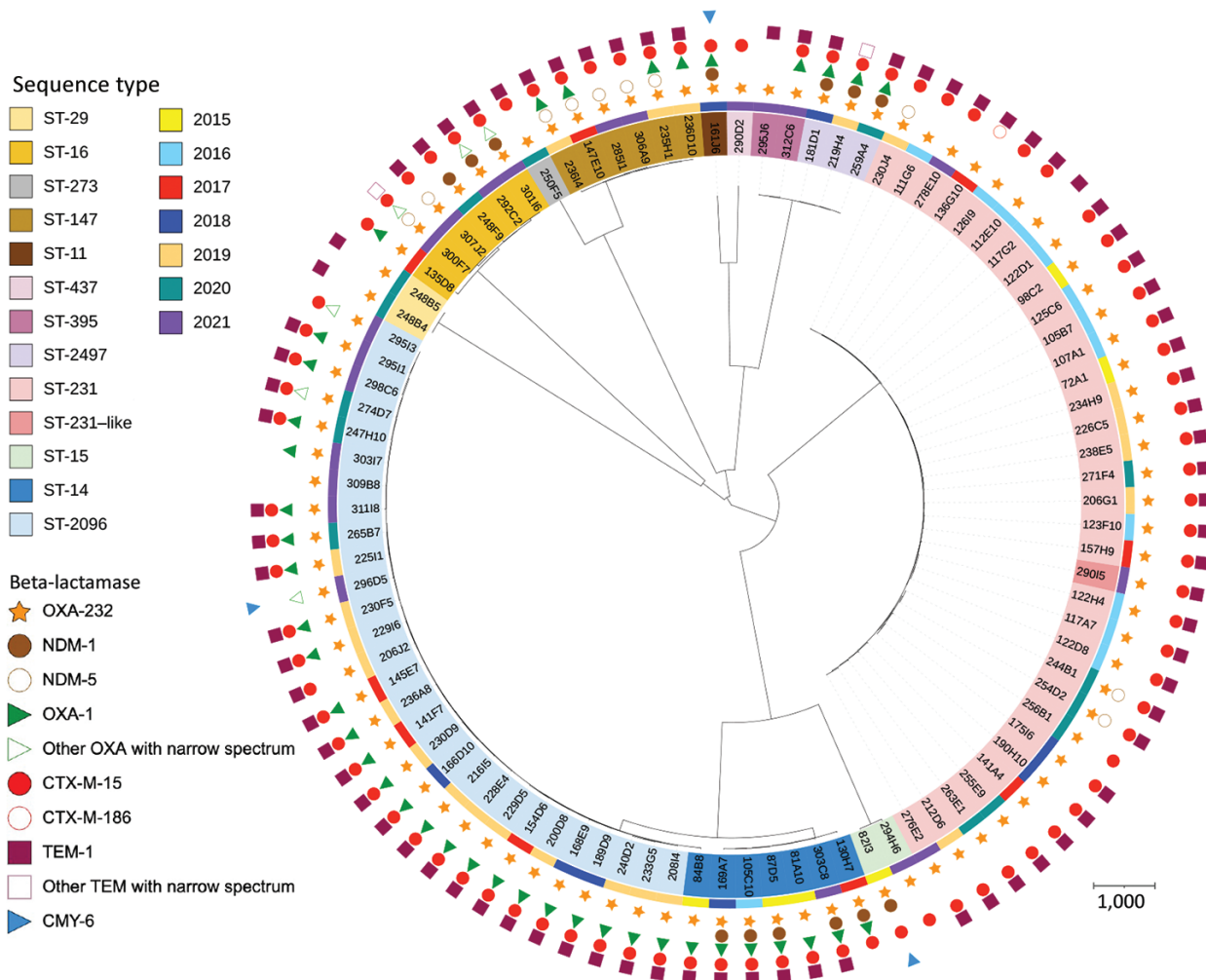
Among the 95 patients colonized or infected with OXA-232–producing *K. pneumoniae*, 19 had recently returned from Asia (including 15 from India) and 12 from the Middle East. Among *K. pneumoniae* isolates, we identified 14 different STs, 5 of which were



**Figure 1.** OXA-232–producing Enterobacteriales received at the National Reference Center for Carbapenem-Resistant Enterobacteriales, France 2013–2021. A) Evolution of several OXA-232–producing Enterobacteriales, by species (top of panel) and carbapenemase variant (bottom). B) Evolution of distribution of ST among all OXA-232–producing *K. pneumoniae*. C) Evolution of distribution of ST among NDM and OXA-232–coproducing *K. pneumoniae*. NDM, New Delhi metallo- $\beta$ -lactamase; OXA, oxacillinase; ST, sequence type.

represented by >5 strains: ST-231 (n = 33), ST-2096 (n = 29), ST-14 (n = 7), ST-16 (n = 6), and ST-147 (n = 6). We observed a diversification in OXA-232-producing *K. pneumoniae* STs over the last 2 years of the study period. In addition, the number of *K. pneumoniae* ST-231 isolates decreased, whereas the number of *K. pneumoniae* ST-2096 isolates increased (Figure 1, panel B). We built single nucleotide polymorphism (SNP) matrices and phylogenetic trees for the 2 main STs (ST-231 and ST-2096) and compared them to epidemiologic data. We considered 2 isolates to be clonally related (probably by cross-transmission) if they differed by <21 SNPs, as previously reported for *K. pneumoniae* clonal complex 258 (15). For both STs, we identified many subclones (20 for ST-231 and 21 for ST-2096) (Figure 2), suggesting polyclonal dissemination including within these 2 high-risk clones.

*K. pneumoniae* coproducing OXA-232 and NDM (NDM-1 or NDM-5) belonged to several STs (ST-14, ST-16, ST-147, ST-231, and ST-2497) but not to ST-2096 (Figure 1, panel C; Figure 2; Appendix Figure). Among the 95 OXA-232-producing *K. pneumoniae*, we identified additional  $\beta$ -lactamases in all strains except 1 (309B8). Eighty-two coproduced Temoniera  $\beta$ -lactamase 1 (32/33 for ST-231 and 25/29 for ST-2096), 86 coproduced the cefotaximase-Munich extended-spectrum  $\beta$ -lactamase 15 (31/33 for ST-231 and 26/29 for ST-2096), and 42 coproduced OXA-1 (0/33 for ST-231 and 25/29 for ST-2096) (Appendix Figure). Furthermore, 3 non-clonally related isolates coproduced the acquired *C. freundii* intrinsic cephalosporinase 6 (ST-231, ST-11, and ST-15) (Appendix Figure). Analysis of the genetic environment revealed that the *bla*<sub>OXA-232</sub> was carried by the 6-kb in size ColE-type plasmid as previously described (5).



**Figure 2.** Phylogenetic relationship of OXA-232-producing *K. pneumoniae* ST-231 (A) and ST-2096 (B) analyzed at the National Reference Center for Carbapenem-Resistant Enterobacteriales, France 2013–2021. The phylogenetic trees were built with an SNP analysis approach. Scale bars under trees indicate the number of SNPs per position of common sequences. OXA, oxacillinase; SNP, single nucleotide polymorphism; ST, sequence type.

## Conclusions

Recent data suggested that the dissemination of OXA-232-producing *K. pneumoniae* is increasing rapidly, especially in Asia and the Middle East (7,11). In our study, about a third of patients had recently visited 1 of these regions. Furthermore, we observed an increasing number of OXA-232 and NDM coproducers. These isolates are of high concern because of their lack of susceptibility to all antimicrobials, including last-resort combinations such as ceftazidime/avibactam, meropenem/vaborbactam, and imipenem/relebactam.

The OXA-232-producing *K. pneumoniae* isolates that are reported to be responsible for outbreaks usually belonged to ST-231, ST-15, ST-16 and ST-147 (4,9). In our study, a wide diversity of STs was found, but the 2 main types were ST-231 and ST-2096. ST-231 was widely reported with OXA-232-producing *K. pneumoniae*, but ST-2096 was first reported only recently in India in 2019 (7,9). ST-2096 in India was also reported to be hypervirulent because it produced characteristic virulence genes such as *rmpA2*, *iutA*, and *iuc* operon (9). Our results suggest that the ST-2096 appeared very recently in France (2017). SNPs analysis demonstrated that the emergence and rapid dissemination of ST-2096 OXA-232-producing *K. pneumoniae* is not linked to a single or a few outbreaks. In our collection, 29 of the 30 ST-2096 *K. pneumoniae* isolates produced OXA-232, whereas the remaining isolate did not produce any carbapenemase, suggesting a recent acquisition of *bla*<sub>OXA-232</sub> in this clone.

A recent publication reported an association between ST-2096 and a higher risk for bacteremia and death (7). In our study, the unique isolate responsible for bacteremia belonged to ST-231. In contrast, 25 of the 29 ST-2096 isolates were cultured from rectal swabs.

As expected, *bla*<sub>OXA-232</sub> was located on a ColE plasmid in all isolates. The close genetic environment of *bla*<sub>OXA-232</sub> involved *ISEcp1* upstream of the *bla*<sub>OXA-232</sub> gene as previously described (5).

## About the Author

Dr. Emeraud is an assistant professor at the Institut National de la Santé et de la Recherche Médicale. Her primary research interests include epidemiology, genetics, and biochemistry of  $\beta$ -lactamases in gram-negative bacteria.

## References

1. Naas T, Dortet L, Iorga BI. Structural and functional aspects of class A carbapenemases. *Curr Drug Targets*. 2016;17:1006–28. <https://doi.org/10.2174/1389450117666160310144501>
2. Mojica MF, Bonomo RA, Fast W. B1-metallo- $\beta$ -lactamases: where do we stand? *Curr Drug Targets*. 2016;17:1029–50. <https://doi.org/10.2174/1389450116666151001105622>
3. Poirel L, Potron A, Nordmann P. OXA-48-like carbapenemases: the phantom menace. *J Antimicrob Chemother*. 2012;67:1597–606. <https://doi.org/10.1093/jac/dks121>
4. Pitout JDD, Peirano G, Kock MM, Strydom KA, Matsumura Y. The global ascendancy of OXA-48-type carbapenemases. *Clin Microbiol Rev*. 2019;33:33. <https://doi.org/10.1128/CMR.00102-19>
5. Potron A, Rondinaud E, Poirel L, Belmonte O, Boyer S, Camiade S, et al. Genetic and biochemical characterisation of OXA-232, a carbapenem-hydrolysing class D  $\beta$ -lactamase from Enterobacteriaceae. *Int J Antimicrob Agents*. 2013;41:325–9. <https://doi.org/10.1016/j.ijantimicag.2012.11.007>
6. Oueslati S, Retaileau P, Marchini L, Berthault C, Dortet L, Bonnín RA, et al. Role of arginine 214 in the substrate specificity of OXA-48. *Antimicrob Agents Chemother*. 2020;64:64. <https://doi.org/10.1128/AAC.02329-19>
7. Isler B, Özer B, Çınar G, Aslan AT, Vatansever C, Falconer C, et al. Characteristics and outcomes of carbapenemase harbouring carbapenem-resistant *Klebsiella* spp. bloodstream infections: a multicentre prospective cohort study in an OXA-48 endemic setting. *Eur J Clin Microbiol Infect Dis*. 2022;41:841–7. <https://doi.org/10.1007/s10096-022-04425-4>
8. Naha S, Sands K, Mukherjee S, Saha B, Dutta S, Basu S. OXA-181-like carbapenemases in *Klebsiella pneumoniae* ST14, ST15, ST23, ST48, and ST231 from septicemic neonates: coexistence with NDM-5, resistome, transmissibility, and genome diversity. *MSphere*. 2021;6:6. <https://doi.org/10.1128/mSphere.01156-20>
9. Shankar C, Mathur P, Venkatesan M, Pragasa AK, Anandan S, Khurana S, et al. Rapidly disseminating *bla*<sub>OXA-232</sub> carrying *Klebsiella pneumoniae* belonging to ST231 in India: multiple and varied mobile genetic elements. *BMC Microbiol*. 2019;19:137. <https://doi.org/10.1186/s12866-019-1513-8>
10. Weng X, Shi Q, Wang S, Shi Y, Sun D, Yu Y. The characterization of OXA-232 carbapenemase-producing ST437 *Klebsiella pneumoniae* in China. *Can J Infect Dis Med Microbiol*. 2020;2020:5626503. <https://doi.org/10.1155/2020/5626503>
11. Zhu Z, Huang H, Xu Y, Wang M, Lv J, Xu L, et al. Emergence and genomics of OXA-232-producing *Klebsiella pneumoniae* in a hospital in Yancheng, China. *J Glob Antimicrob Resist*. 2021;26:194–8. <https://doi.org/10.1016/j.jgar.2021.05.015>
12. Avolio M, Vignaroli C, Crapis M, Camporese A. Co-production of NDM-1 and OXA-232 by ST16 *Klebsiella pneumoniae*, Italy, 2016. *Future Microbiol*. 2017;12:1119–22. <https://doi.org/10.2217/fmb-2017-0041>
13. Doi Y, Hazen TH, Boitano M, Tsai YC, Clark TA, Korlach J, et al. Whole-genome assembly of *Klebsiella pneumoniae* coproducing NDM-1 and OXA-232 carbapenemases using single-molecule, real-time sequencing. *Antimicrob Agents Chemother*. 2014;58:5947–53. <https://doi.org/10.1128/AAC.03180-14>
14. Kesaramangalam Kalyanavenkatramanan S, Sewunet T, Wangchinda W, Tangkoskul T, Thamlikitkul V, Giske CG, et al. Optical DNA mapping of plasmids reveals clonal spread of carbapenem-resistant *Klebsiella pneumoniae* in a large Thai hospital. *Antibiotics (Basel)*. 2021;10:10. <https://doi.org/10.3390/antibiotics10091029>
15. David S, Reuter S, Harris SR, Glasner C, Feltwell T, Argimon S, et al.; EuSCAPE Working Group; ESGEM Study Group. Epidemic of carbapenem-resistant *Klebsiella pneumoniae* in Europe is driven by nosocomial spread. *Nat Microbiol*. 2019;4:1919–29. <https://doi.org/10.1038/s41564-019-0492-8>

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## Appendix

**Appendix Table 1.** OXA-232-producing isolates included in the study

Strain	Species	Year	Origin	Carbapenemase	ST	Genome Accession Number
72A1	<i>K. pneumoniae</i>	2015	Rectal swab	OXA-232	231	JALXFX000000000
81A10	<i>K. pneumoniae</i>	2015	Rectal swab	OXA-232	14	JALXFY000000000
82I3	<i>K. pneumoniae</i>	2015	Rectal swab	OXA-232 + NDM-1	15	JALXFZ000000000
84B8	<i>K. pneumoniae</i>	2015	Rectal swab	OXA-232	14	JALXGA000000000
87D5	<i>K. pneumoniae</i>	2015	Rectal swab	OXA-232 + NDM-1	14	JALXGB000000000
98C2	<i>K. pneumoniae</i>	2015	Rectal swab	OXA-232	231	JALXFW000000000
105B7	<i>K. pneumoniae</i>	2016	Blood culture	OXA-232	231	JALXGC000000000
105C10	<i>K. pneumoniae</i>	2016	Rectal swab	OXA-232 + NDM-1	14	JALXGD000000000
107A1	<i>K. pneumoniae</i>	2016	Rectal swab	OXA-232	231	JALXGE000000000
111G6	<i>K. pneumoniae</i>	2016	Other or unknown origin	OXA-232	231	JALXGF000000000
112E10	<i>K. pneumoniae</i>	2016	Rectal swab	OXA-232	231	JALXGG000000000
117A7	<i>K. pneumoniae</i>	2016	Respiratory tract	OXA-232	231	JALXGH000000000
117G2	<i>K. pneumoniae</i>	2016	Rectal swab	OXA-232	231	JALXGI000000000
122D1	<i>K. pneumoniae</i>	2016	Rectal swab	OXA-232	231	JALXGJ000000000
122D8	<i>K. pneumoniae</i>	2016	Rectal swab	OXA-232	231	JALXGK000000000
122H4	<i>K. pneumoniae</i>	2016	Rectal swab	OXA-232	231	JALXGL000000000
123F10	<i>K. pneumoniae</i>	2016	Rectal swab	OXA-232	231	JALXGM000000000
125C6	<i>K. pneumoniae</i>	2016	Rectal swab	OXA-232	231	JALXGN000000000
126I9	<i>K. pneumoniae</i>	2016	Rectal swab	OXA-232	231	JALXGO000000000
130H7	<i>K. pneumoniae</i>	2017	Rectal swab	OXA-232 + NDM-1	14	JALXGP000000000
135D8	<i>K. pneumoniae</i>	2017	Rectal swab	OXA-232	16	JALXGQ000000000
136G10	<i>K. pneumoniae</i>	2017	Rectal swab	OXA-232	231	JALXGR000000000
141A4	<i>K. pneumoniae</i>	2017	Other or unknown origin	OXA-232	231	JALXGS000000000
141F7	<i>K. pneumoniae</i>	2017	Rectal swab	OXA-232	2096	JALXGT000000000
145E7	<i>K. pneumoniae</i>	2017	Other or unknown origin	OXA-232	2096	JANJFV000000000
147E10	<i>K. pneumoniae</i>	2017	Rectal swab	OXA-232 + NDM-5	147	JALXGU000000000
154D6	<i>K. pneumoniae</i>	2017	Rectal swab	OXA-232	2096	JALXGV000000000
157H9	<i>K. pneumoniae</i>	2017	Urines	OXA-232	231	JALXGW000000000
161J6	<i>K. pneumoniae</i>	2018	Rectal swab	OXA-232 + NDM-1	11	JALXGX000000000
166D10	<i>K. pneumoniae</i>	2018	Urines	OXA-232	2096	JALXGY000000000
168E9	<i>K. pneumoniae</i>	2018	Rectal swab	OXA-232	2096	JALXGZ000000000
169A7	<i>K. pneumoniae</i>	2018	Rectal swab	OXA-232 + NDM-1	14	JALXHA000000000

Strain	Species	Year	Origin	Carbapenemase	ST	Genome Accession Number
175I6	<i>K. pneumoniae</i>	2018	Rectal swab	OXA-232	231	JALXHB000000000
181D1	<i>K. pneumoniae</i>	2018	Urines	OXA-232 + NDM-1	2497	JALXHC000000000
189D9	<i>K. pneumoniae</i>	2018	Rectal swab	OXA-232	2096	JANJFW000000000
190H10	<i>K. pneumoniae</i>	2018	Rectal swab	OXA-232	231	JALXHD000000000
200D8	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JALXHE000000000
206G1	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	231	JALXHF000000000
206J2	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JALXHG000000000
208I4	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JALXHH000000000
212D6	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	231	JALXHI000000000
216I5	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JALXHJ000000000
219H4	<i>K. pneumoniae</i>	2019	Urines	OXA-232 + NDM-1	2497	JALXHK000000000
225I1	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JALXHL000000000
226C5	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	231	JALXHM000000000
228E4	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JALXHN000000000
229D5	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JALXHO000000000
229I6	<i>K. pneumoniae</i>	2019	Urines	OXA-232	2096	JALXHP000000000
230D9	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JANJFX000000000
230F5	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JALXHQ000000000
230J4	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	231	JALXHR000000000
233G5	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JANJFY000000000
234H9	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	231	JALXHS000000000
235H1	<i>K. pneumoniae</i>	2019	Urines	OXA-232	147	JALXHT000000000
236A8	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JALXHU000000000
236D10	<i>K. pneumoniae</i>	2019	Urines	OXA-232 + NDM-5	147	JALXHV000000000
236I4	<i>K. pneumoniae</i>	2019	Urines	OXA-232 + NDM-5	147	JALXHW000000000
238E5	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	231	JALXHX000000000
240D2	<i>K. pneumoniae</i>	2019	Rectal swab	OXA-232	2096	JALXHY000000000
244B1	<i>K. pneumoniae</i>	2020	Rectal swab	OXA-232 + NDM-5	231	JALXHZ000000000
247H10	<i>K. pneumoniae</i>	2020	Urines	OXA-232	2096	JANJFZ000000000
248B4	<i>K. pneumoniae</i>	2020	Rectal swab	OXA-232	29	JALXIA000000000
248B5	<i>K. pneumoniae</i>	2020	Rectal swab	OXA-232	29	JALXIB000000000
248F9	<i>K. pneumoniae</i>	2020	Urines	OXA-232 + NDM-1	16	JALXIC000000000
250F5	<i>K. pneumoniae</i>	2020	Rectal swab	OXA-232	273	JALXID000000000
254D2	<i>K. pneumoniae</i>	2020	Rectal swab	OXA-232 + NDM-5	231	JALXIE000000000
255E9	<i>K. pneumoniae</i>	2020	Other or unknown origin	OXA-232	231	JALXIF000000000
256B1	<i>K. pneumoniae</i>	2020	Rectal swab	OXA-232	231	JALXIG000000000
259A4	<i>K. pneumoniae</i>	2020	Urines	OXA-232 + NDM-1	2497	JALXIH000000000
263E1	<i>K. pneumoniae</i>	2020	Rectal swab	OXA-232	231	JALXII000000000
265B7	<i>K. pneumoniae</i>	2020	Rectal swab	OXA-232	2096	JALXIJ000000000
271F4	<i>K. pneumoniae</i>	2020	Other or unknown origin	OXA-232	231	JALXIK000000000
274D7	<i>K. pneumoniae</i>	2020	Rectal swab	OXA-232	2096	JALXIL000000000
276E2	<i>K. pneumoniae</i>	2021	Urines	OXA-232	231	JALXIM000000000
278E10	<i>K. pneumoniae</i>	2021	Urines	OXA-232	231	JALXIN000000000
285I1	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232 + NDM-5	147	JALXIO000000000
290D2	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232	437	JALXIP000000000
290I5	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232	New	JALXIQ000000000
292C2	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232 + NDM-1	16	JALXIR000000000

Strain	Species	Year	Origin	Carbapenemase	ST	Genome Accession Number
294H6	<i>K. pneumoniae</i>	2021	Other or unknown origin	OXA-232	15	JALXIS000000000
295I1	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232	2096	JALXIT000000000
295I3	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232	2096	JALXIU000000000
295J6	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232	395	JALXIV000000000
296D5	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232	2096	JALXIW000000000
298C6	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232	2096	JALXIX000000000
300F7	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232 + NDM-5	16	JALXIY000000000
301I6	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232 + NDM-1	16	JALXIZ000000000
303C8	<i>K. pneumoniae</i>	2021	Urines	OXA-232 + NDM-1	14	JALXJA000000000
303I7	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232	2096	JALXJB000000000
304B3	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232	512	JALXJC000000000
306A9	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232 + NDM-5	147	JALXJD000000000
307J2	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232 + NDM-5	16	JALXJE000000000
309B8	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232	2096	JALXJF000000000
311I8	<i>K. pneumoniae</i>	2021	Rectal swab	OXA-232	2096	JALXJG000000000
312C6	<i>K. pneumoniae</i>	2021	Urines	OXA-232	395	JALXJH000000000
ND	<i>E. coli</i>	2013	Rectal swab	OXA-232 + NDM-1	ND	
ND	<i>K. pneumoniae</i>	2013	Rectal swab	OXA-232 + NDM-1	ND	
ND	<i>K. pneumoniae</i>	2013	Rectal swab	OXA-232 + NDM-1	ND	
43B6	<i>K. pneumoniae</i>	2014	Rectal swab	OXA-232	ND	
44C5	<i>K. pneumoniae</i>	2014	Rectal swab	OXA-232	ND	
46A8	<i>E. coli</i>	2014	Other or unknown origin	OXA-232 + NDM-1	ND	
68B2	<i>E. coli</i>	2014	Other or unknown origin	OXA-232	ND	
111C6	<i>E. coli</i>	2016	Rectal swab	OXA-232	ND	
117C6	<i>E. coli</i>	2016	Rectal swab	OXA-232	ND	
148H7	<i>E. coli</i>	2017	Rectal swab	OXA-232 + NDM-5	ND	
227H1	<i>E. coli</i>	2019	Rectal swab	OXA-232	ND	
165B3	<i>C. freundii</i>	2018	Rectal swab	OXA-232	ND	
170F5	<i>C. freundii</i>	2018	Urines	OXA-232	ND	
188H3	<i>C. freundii</i>	2018	Urines	OXA-232	ND	
203I10	<i>C. freundii</i>	2019	Rectal swab	OXA-232	ND	
207F9	<i>C. freundii</i>	2019	Rectal swab	OXA-232 + NDM-1	ND	
213I9	<i>C. freundii</i>	2019	Rectal swab	OXA-232	ND	
229I3	<i>C. freundii</i>	2019	Rectal swab	OXA-232	ND	
230B3	<i>C. freundii</i>	2019	Rectal swab	OXA-232	ND	
236A9	<i>C. freundii</i>	2019	Rectal swab	OXA-232	ND	
263J3	<i>C. freundii</i>	2020	Rectal swab	OXA-232 + NDM-5	ND	
285D4	<i>C. freundii</i>	2021	Urines	OXA-232 + NDM-1	ND	
292G1	<i>C. freundii</i>	2021	Rectal swab	OXA-232	ND	
299H2	<i>C. freundii</i>	2021	Other or unknown origin	OXA-232	ND	
214J10	<i>K. aerogenes</i>	2019	Rectal swab	OXA-232	ND	
255E10	<i>K. aerogenes</i>	2020	Blood culture	OXA-232	ND	
85F5	<i>K. oxytoca</i>	2015	Rectal swab	OXA-232	ND	

ST, sequence type obtained by MLST; ND, not determined.

**Appendix Table 2.** Percentage of OXA-232 producing isolates received at the French National Reference Center for carbapenem-resistant Enterobacterales among all carbapenemase-producing Enterobacterales (CPE) or among OXA-48-like from 2013 to 2021.

Year	OXA-232 and OXA-232 + NDM (n)	% among CPE	% among OXA-48 like	OXA-232 (n)	% among CPE	% among OXA-48 like	OXA-232 + NDM (n)	% among CPE	% among OXA-48 like
2013	3	0.47%	0.59%	0	0.00%	0.00%	3	0.47%	0.59%
2014	4	0.37%	0.43%	3	0.28%	0.33%	1	0.09%	0.11%
2015	7	0.55%	0.71%	5	0.39%	0.51%	2	0.16%	0.20%
2016	15	0.97%	1.26%	14	0.90%	1.18%	1	0.06%	0.08%
2017	10	0.52%	0.69%	8	0.42%	0.56%	2	0.10%	0.14%
2018	10	0.37%	0.52%	8	0.30%	0.42%	2	0.07%	0.10%
2019	30	0.99%	1.40%	26	0.86%	1.22%	4	0.13%	0.19%
2020	16	0.73%	1.14%	12	0.54%	0.86%	4	0.18%	0.29%
2021	23	0.94%	1.40%	17	0.69%	1.03%	6	0.25%	0.36%
2013–2018	49	0.54%	0.70%	38	0.42%	0.55%	11	0.12%	0.16%
2019–2021	69	0.91%	1.33%	55	0.72%	1.06%	14	0.18%	0.27%

**Appendix Figure.** Global characterization (sequence type, year of isolation,  $\beta$ -lactamase content) of nonduplicate 95 OXA-232–producing *Klebsiella pneumoniae* analyzed at the National Reference Center for Carbapenem-Resistant Enterobacterales, France, 2013–2021. Scale bar indicates the number of SNP per position of common sequences. CMY-6, variant of *C. freundii* intrinsic cephalosporinase; CTX-M, cefotaximase–Munich extended-spectrum  $\beta$ -lactamase; OXA, oxacillinase; NDM, New Delhi metallo- $\beta$ -lactamase; ST, sequence type, TEM, Temoniera  $\beta$ -lactamase.