

Legends to supplementary materials

Dataset S1. *Klebsiella* genomic data used in this study. Part A, *Klebsiella* genomes retrieved from the NCBI Genome database. Part B, chromosomes and plasmids in the complete *Klebsiella* genomes. Part C, other *Klebsiella* plasmids retrieved from the NCBI RefSeq database.

Dataset S2. *In silico* IncFII_K typing results in *Klebsiella* genomes.

Dataset S3. *In silico* multilocus sequence typing (MLST) of *Klebsiella pneumoniae*

Dataset S4. The IncFII_K plasmids with full sequences analysed in this study.

Dataset S5. The antibiotic resistance genes identified on IncFII_K plasmids.

Dataset S6. The transfer regions of IncFII_K plasmids.

Table S1. Distribution of IncFII_K replicons found in the *Klebsiella* genomes.

Table S2. Distribution of multi-replicon IncFII_K plasmids.

Figure S1. The IncFII_K alleles and PlasmidFinder probes used for *in silico* IncFII_K plasmid typing. Alleles were downloaded from pMLST. Colours indicated different allele groups. Percentages in brackets are identities in nucleotide levels between the alleles and the best matching probes. Multiple sequence alignment was performed by MUSCLE v3.7. Then the tree was constructed using neighbor-joining (NJ) method (A) and maximum likelihood (ML) method (B) with 1,000 bootstrap replicates in MEGA5. The two trees display similar topology. IncFII_K alleles (K1-K17) from pMLST were used to fish out the IncFII_K replicon probes from PlasmidFinder. The alleles K1-K3, K5-K8, K11, and K13-16 matched to the literally IncFII_K probe “IncFII(K)_1__CP000648” as the best hit with 95%-100% BLASTn identities. In contrast, alleles K4, K9, K10, K12 and K17 matched to probe

“IncFII_1_pKP91_CP000966” as the best hit. Alleles K4 and K9 were 96% - 100% identical to the probe sequence, while K10, K12 and K17 displayed a lower identity (88%-93%). Although there is literal discrepancy in name, the probe “IncFII_1_pKP91_CP000966” was defined by plasmid pKP91 (GenBank accession number: CP000966), which is also an IncFII_K plasmid.

Figure S2. The *in silico* IncFII_K plasmid typing strategy.

Figure S3. Neighbor-joining (NJ) tree and maximum likelihood (ML) trees of all IncFII_K alleles. The tree was constructed using NJ method (left) and ML method (rightB) with 1,000 bootstrap replicates in MEGA5. The two trees display similar topology. Colours indicated different allele groups. Percentages in brackets are identities in nucleotide levels between the alleles and the best matching probes.

Figure S4. Plasmids of same IncFII_K type are widespread in *K. pneumoniae* with different sequence types. Histogram is the count of occurrence of the IncFII_K alleles in *Klebsiella pneumoniae* genomes specified by sequence type. In the block is the zoom-in view of types IncFII_{K3}-IncFII_{K33} and unclassified alleles.

Figure S5. Comparison of the conjugation modules between plasmid pKPHS2 and plasmid F. GenBank accession numbers are CP003224 for pKPHS2 and AP001918 for plasmid F. Gene organisation is drawn to scale. Homologous counterparts are connected by orange lines. Identities in amino acid level of the components between the two plasmids are shown below.

Figure S5. Phylogenetic tree of the DNA sequences of the transfer regions of the studied plasmids. Plasmids with same names are discriminated by their accession numbers in the

brackets. The letters 'a' and 'b' are used to distinguish the two transfer regions on plasmid pKPC-727. Multiple sequence alignment was performed by Kalign v2.04. Then the tree was constructed using neighbor-joining method with 1,000 bootstrap replicates in MEGA5.

Figure S7. Phylogenetic trees of the proteins TraV and TrbE and TrbF of the studied plasmids. Plasmids with same names are discriminated by their accession numbers in the brackets. The letters 'a' and 'b' are used to distinguish the two conjugation modules on plasmid pKPC-727. Multiple sequence alignment was performed by MUSCLE v3.7. Then the tree was constructed using neighbor-joining method with 1,000 bootstrap replicates in MEGA5.

Table S1. Distribution of IncFII_K replicons found in the *Klebsiella* genomes.

Species	Analysed genome no.	No. of genomes containing IncFII _K	Positive ratio	IncFII _K replicons no.	No. of IncFII _K replicons on plasmids	No. of IncFII _K replicons on contigs
<i>K. cf. planticola</i> B43	1					
<i>K. michiganensis</i>	46	12	26.09%	12	1	11
<i>K. oxytoca</i>	84	16	19.05%	16	1	15
<i>K. pneumoniae</i>	1,258	868	69.00%	1,014	40	974
<i>K. quasipneumoniae</i>	15	5	33.33%	5	1	4
<i>Klebsiella</i> sp. 10982	1	1	100.00%	1		1
<i>Klebsiella</i> sp. 1 1 55	1					
<i>Klebsiella</i> sp. 4 1 44FAA	1	1	100.00%	2		2
<i>Klebsiella</i> sp. AA405	1					
<i>Klebsiella</i> sp. AS10	1					

<i>Klebsiella</i> sp. D5A	1	1	100.00%	2		2
<i>Klebsiella</i> sp. G5	1					
<i>Klebsiella</i> sp. KGM-IMP216	1	1	100.00%	1		1
<i>Klebsiella</i> sp. KTE92	1					
<i>Klebsiella</i> sp. MS 92-3	1	1	100.00%	1		1
<i>Klebsiella</i> sp. OBRC7	1					
<i>Klebsiella</i> sp. RIT-PI-d	1					
<i>Klebsiella</i> sp. S1	1					
<i>K. variicola</i>	24	12	50%	15	1	14
<i>Klebsiella variicola</i>	0					
CAG:634						
Total	1,441	918	63.71%	1,069	44	1,025

Data for *K. pneumoniae* are in bold.

Grey lettering indicates the species not analysed.

IncFII_K replicons are listed in Supplementary Dataset S3.

IncFII _{K13}			1									1
IncFII _{K14}						2						2
IncFII _{K2}			2									2
IncFII _{K21}					1							1
IncFII _{K25}			1									1
IncFII _{K17}	13											1
Total	8	6	28	22	1	6	2	1	1	1	1	77

Detail information is listed in Supplementary Dataset S4.

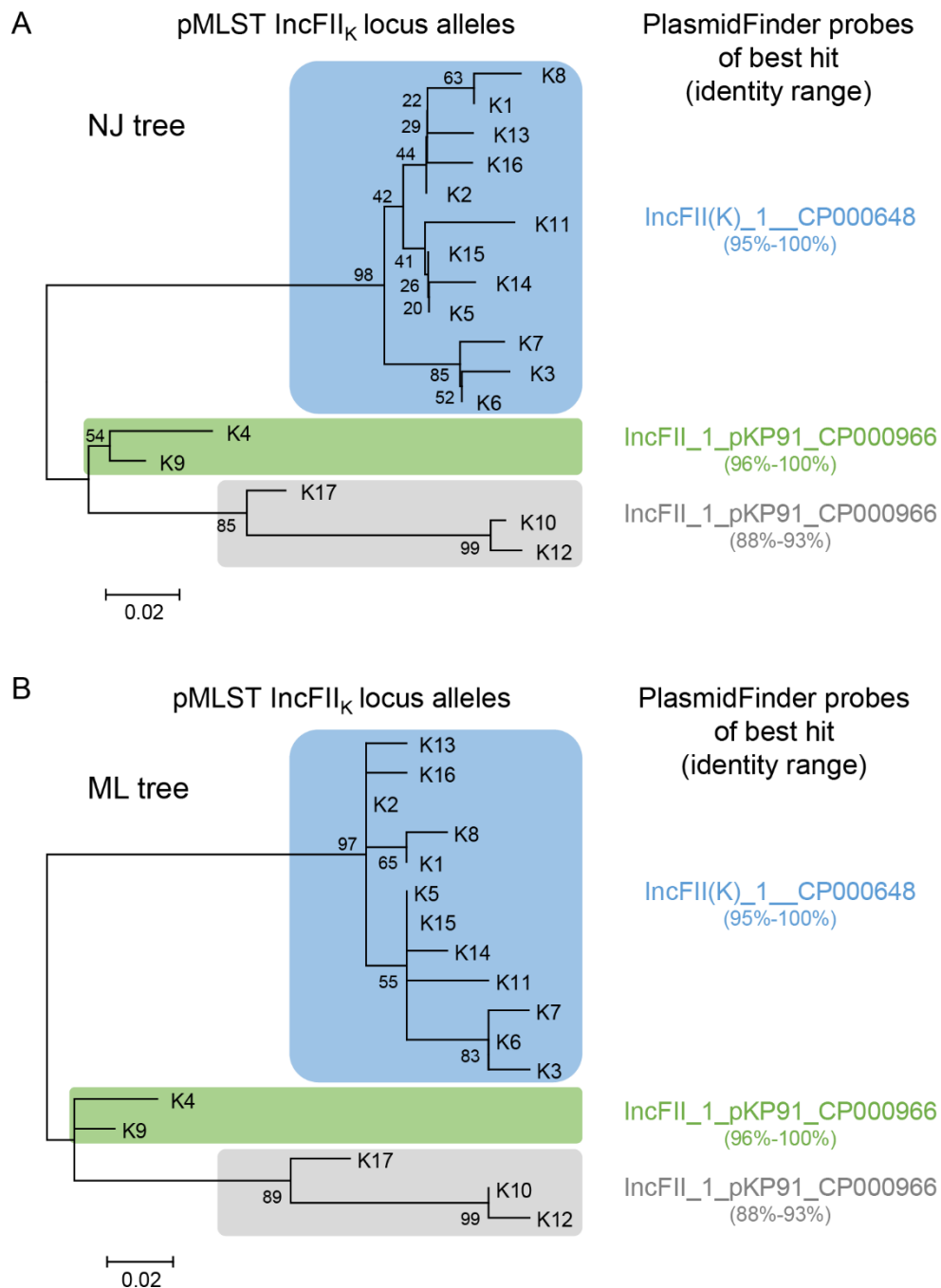


Figure S1. The IncFII_K alleles and PlasmidFinder probes used for *in silico* IncFII_K plasmid typing. Alleles were downloaded from pMLST. Colours indicated different allele groups. Percentages in brackets are identities in nucleotide levels between the alleles and the best matching probes. Multiple sequence alignment was performed by MUSCLE v3.7. Then the tree was constructed using neighbor-joining (NJ) method (A) and maximum likelihood (ML) method (B) with 1,000 bootstrap replicates in MEGA5. The two trees display similar

topology. IncFII_K alleles (K1-K17) from pMLST were used to fish out the IncFII_K replicon probes from PlasmidFinder. The alleles K1-K3, K5-K8, K11, and K13-16 matched to the literally IncFII_K probe “IncFII(K)_1__CP000648” as the best hit with 95%-100% BLASTn identities. In contrast, alleles K4, K9, K10, K12 and K17 matched to probe “IncFII_1_pKP91_CP000966” as the best hit. Alleles K4 and K9 were 96% - 100% identical to the probe sequence, while K10, K12 and K17 displayed a lower identity (88%-93%). Although there is literal discrepancy in name, the probe “IncFII_1_pKP91_CP000966” was defined by plasmid pKP91 (GenBank accession number: CP000966), which is also an IncFII_K plasmid. The IncFII_K alleles fall into three groups according to the probe-mapping profile and the grouping was supported by phylogenetic analysis of those alleles. Therefore, we used the two probes and the 17 alleles to define IncFII_K plasmids and adopted a customised set of thresholds.

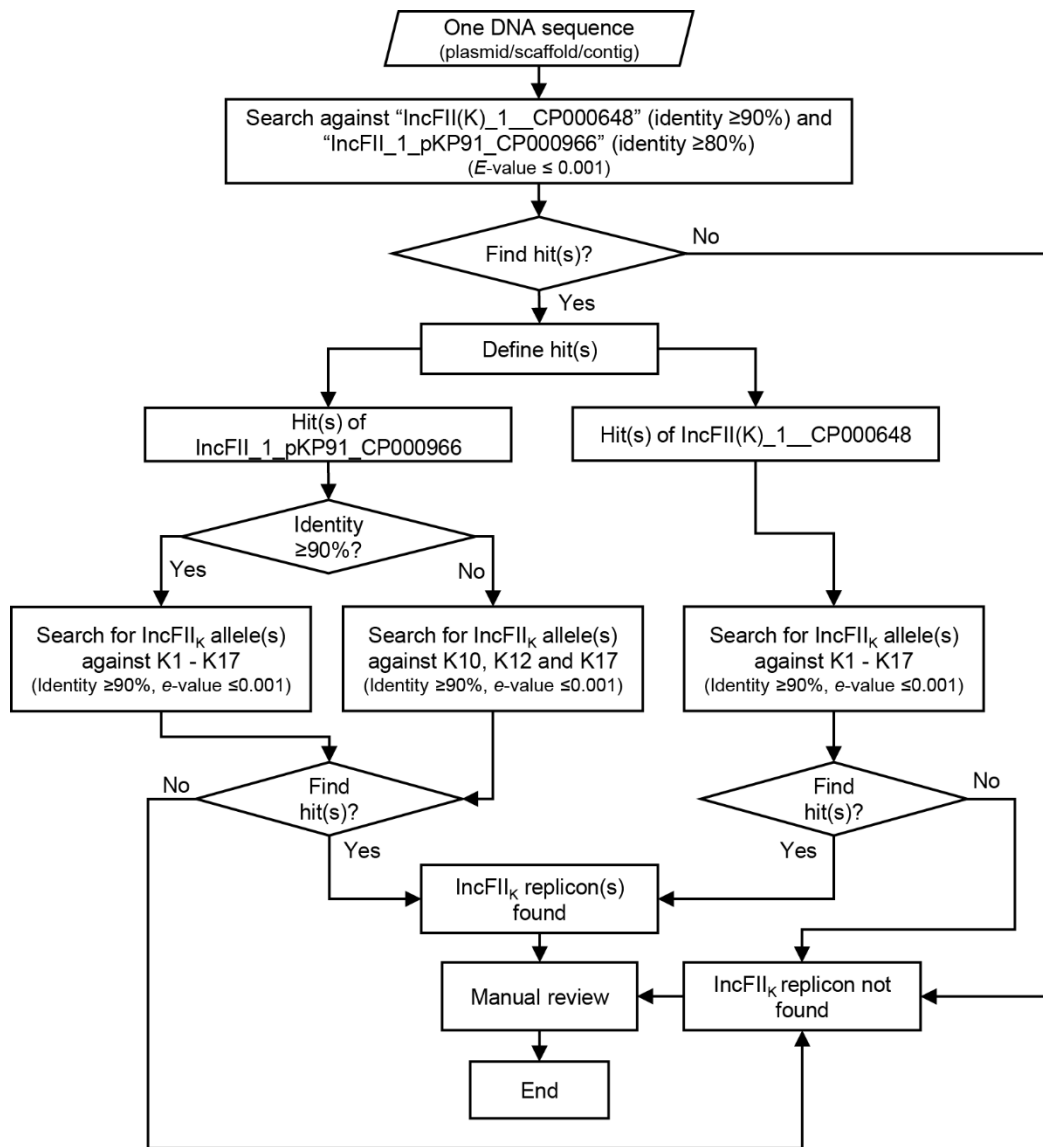


Figure S2. The *in silico* IncFII_K plasmid typing strategy.

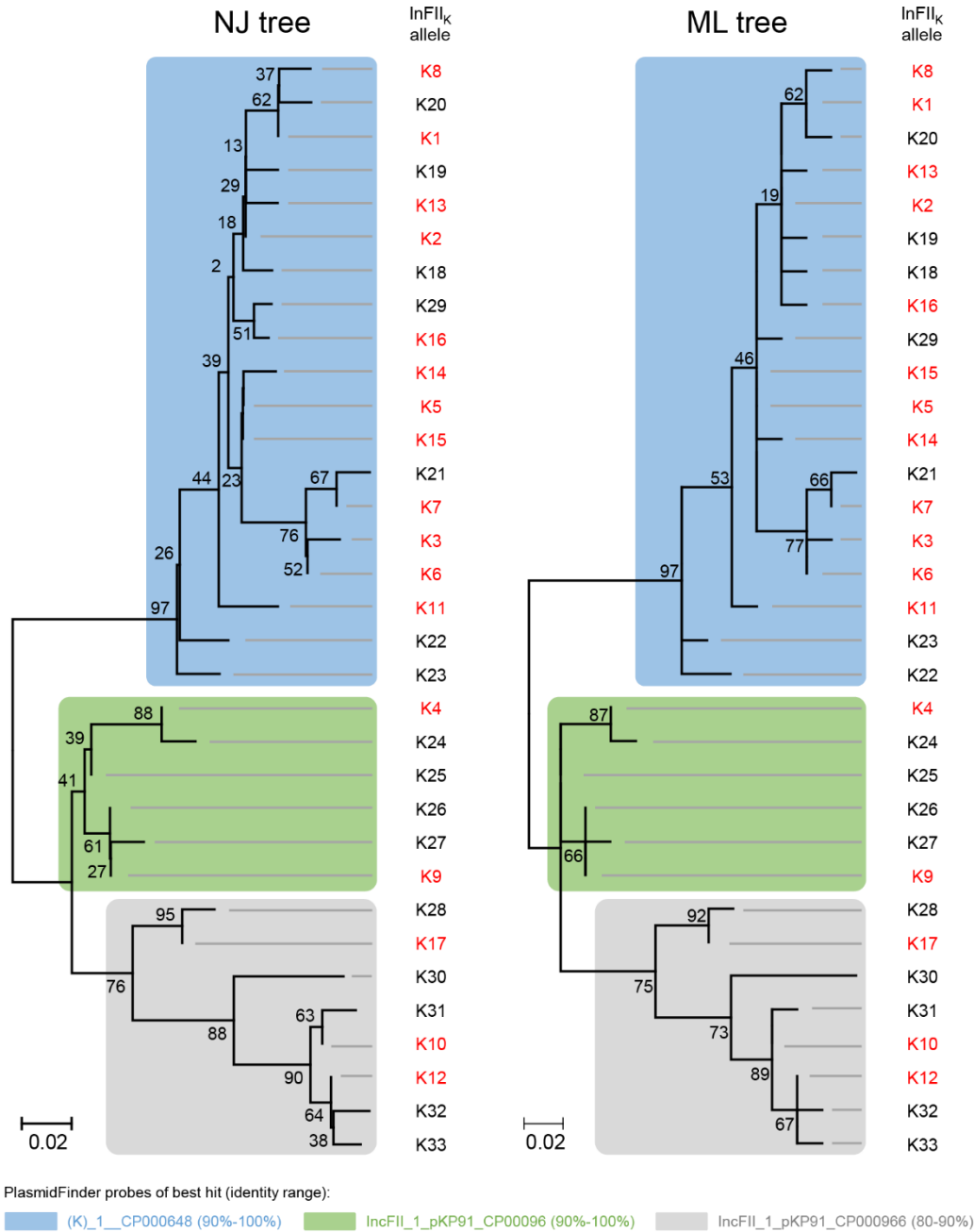


Figure S3. Neighbor-joining (NJ) tree and maximum likelihood (ML) trees of all IncFII_K alleles. The tree was constructed using NJ method (left) and ML method (right) with 1,000 bootstrap replicates in MEGA5. The two trees display similar topology. Colours indicated different allele groups. Percentages in brackets are identities in nucleotide levels between the alleles and the best matching probes.

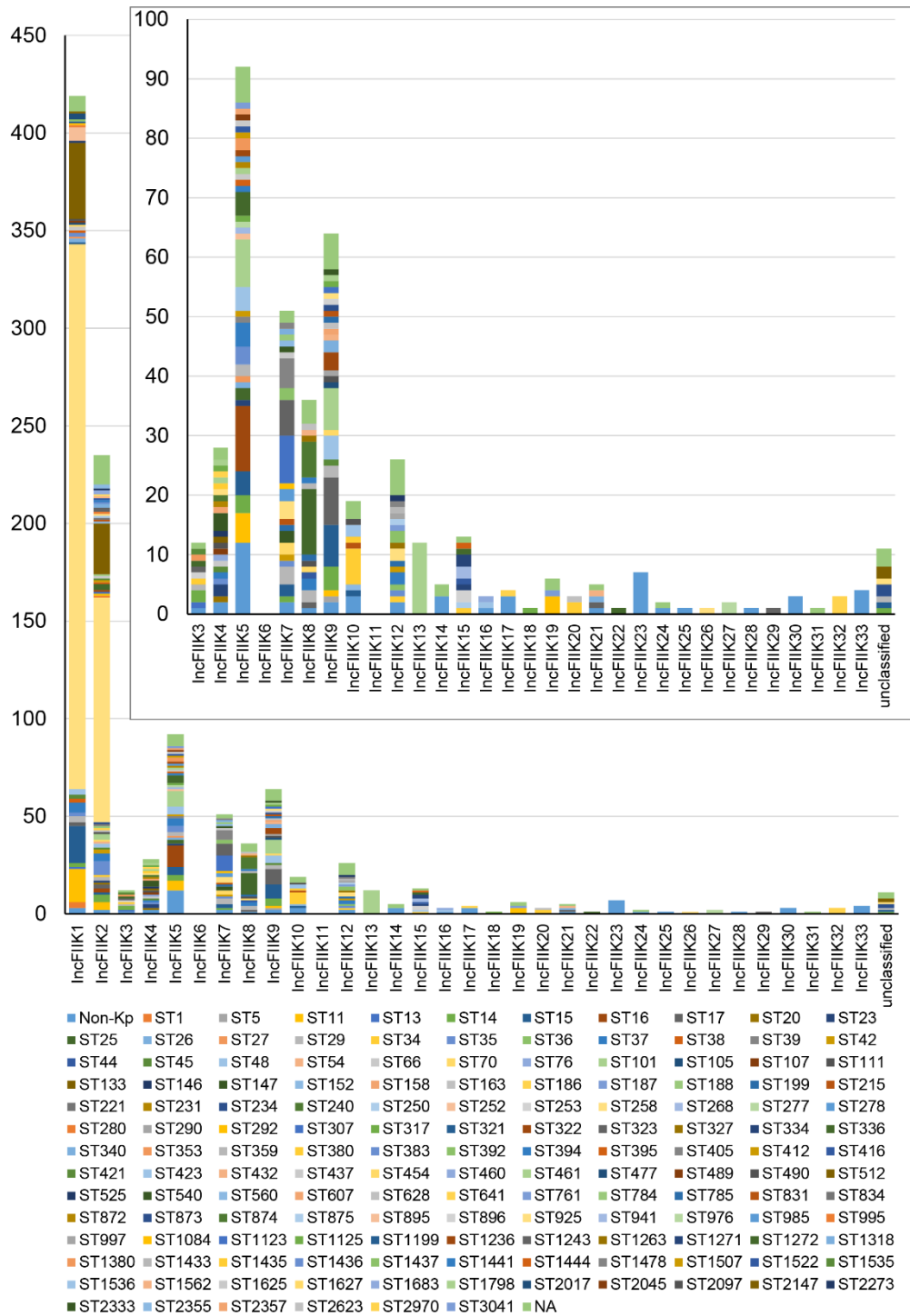


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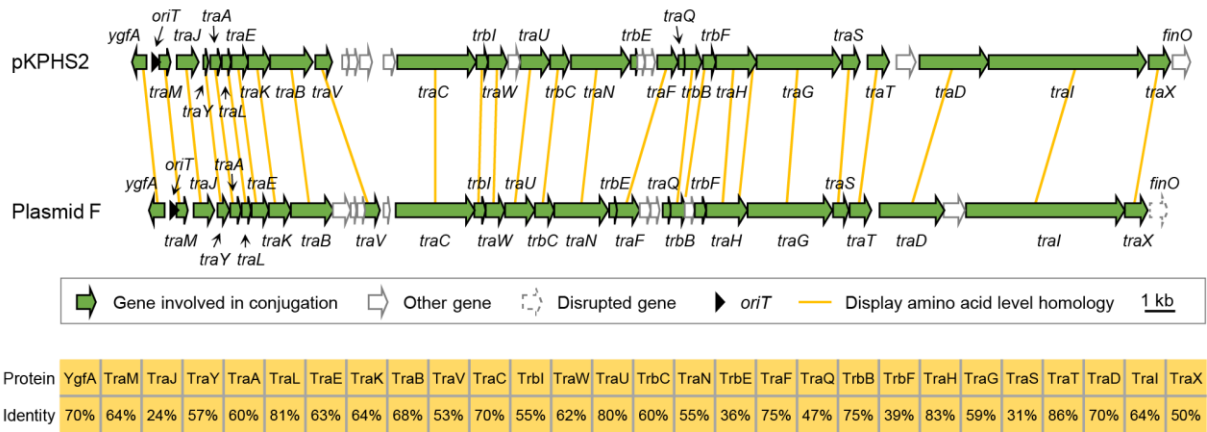


Figure S5. Comparison of the conjugation modules between plasmid pKPHS2 and plasmid F. GenBank accession numbers are CP003224 for pKPHS2 and AP001918 for plasmid F. Gene organisation is drawn to scale. Homologous counterparts are connected by orange lines. Identities in amino acid level of the components between the two plasmids are shown below.

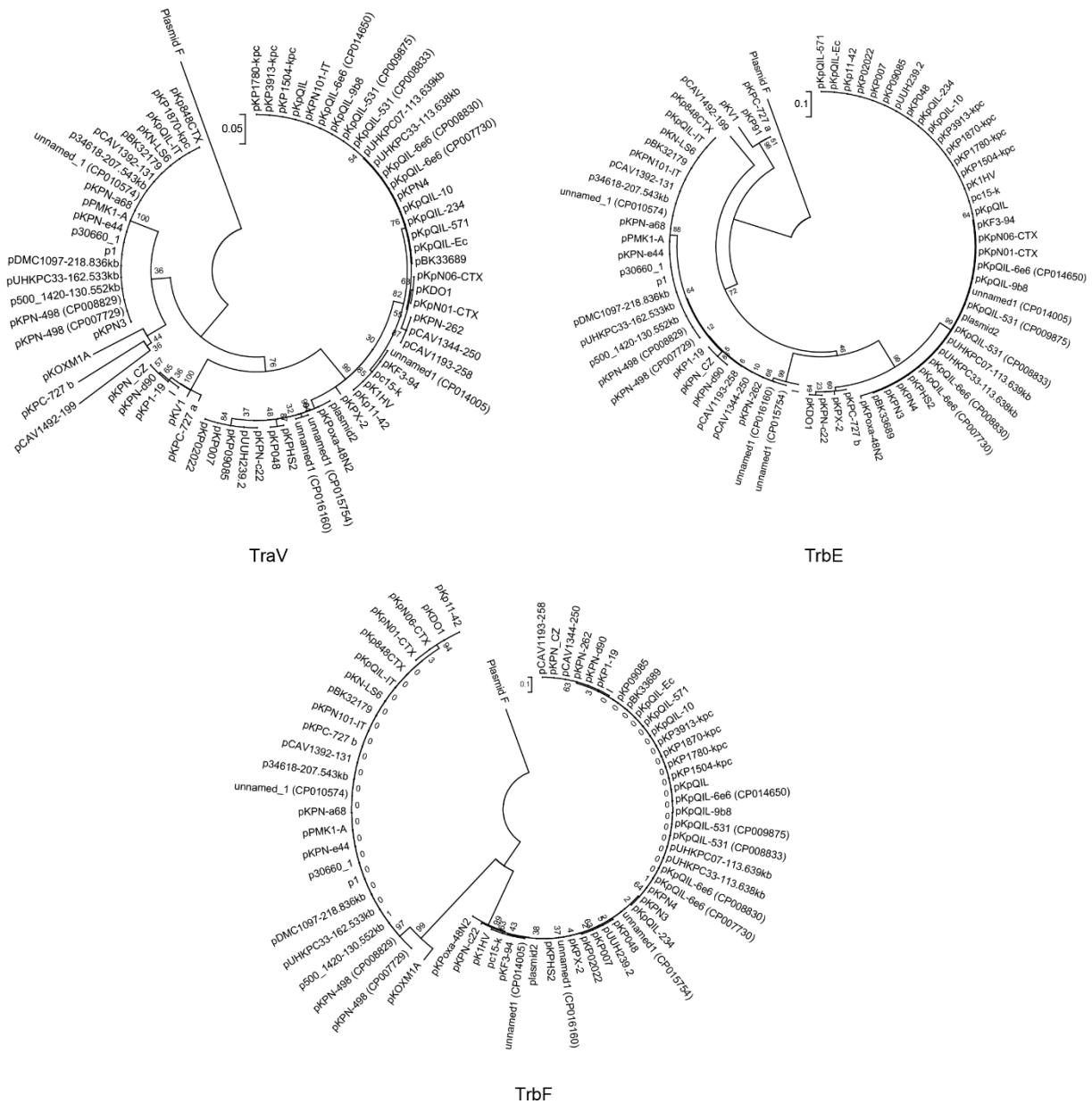


Figure S7. Phylogenetic trees of the proteins TraV and TrbE and TrbF of the studied plasmids. Plasmids with same names are discriminated by their accession numbers in the brackets. The letters ‘a’ and ‘b’ are used to distinguish the two conjugation modules on plasmid pKPC-727. Multiple sequence alignment was performed by MUSCLE v3.7. Then the tree was constructed using neighbor-joining method with 1,000 bootstrap replicates in MEGA5.