

Supplementary Materials for

Characterization of Carbapenem-Resistant *Klebsiella pneumoniae* in Bloodstream Infections: Antibiotic Resistance, Virulence, and Treatment Strategies

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Table S1 Primers for carbapenem-resistant genes detection.

Gene names	Primer sequences (5'-3')	Length (bp)
KPC	F: CGTCTAGTTCTGCTGTCTTG	798
	R: CTTGTCATCCTTGTTAGGCG	
NDM	F: GGTTTGGCGATCTGGTTTTTC	621
	R: CGGGAATGGCTCATCACGATC	
VEB	F: ATGCAAAGCGTTATGAAATTTCCG	553
	R: CTGCTGCAATTCCATTTATTTATCC	
OXA-48	F: GCGTGGTTAAGGATGAACAC	438
	R: CATCAAGTTCAACCCAACCG	
GES	F: ATGCGCTTCATTACGCAC	864
	R: CTATTTGTCCGTGCTCAGG	
IMP	F: GGAATAGAGTGGCTTAAYTCTC	232
	R: GGTTTAAYAAAACAACCACC	
VIM	F: ATGTTAAAAGTTATTAGTAGT	801
	R: CTACTCGGCGACTGAGCGAT	

Note: F represents the forward primer, and R represents the reverse primer.

Primers mentioned above for carbapenem-resistant genes detection were designed from the reference (1).

Table S2 Detailed PCR reaction conditions for carbapenem-resistant genes detection.

Reaction stage	Reaction temperature	Reaction time	Cycle numbers
Initial degeneration	95°C	6 min	1
Denaturation	95°C	30 s	
Annealing	X	30 s	30
Extension	72°C	Y	
Final extension	72°C	7 min	1

Note: PCR conditions include initial denaturation, denaturation, annealing, and extension steps with specified temperatures and durations. The annealing temperature X and extension time Y were adjusted according to the sequence and fragment length of the primers for each gene.

Table S3 Primers for virulence-associated genes detection.

Name of genes	Primer sequence (5'-3')	Length (bp)
<i>K1</i>	F: GGTGCTCTTTACATCATTGC R: GCAATGGCCATTTGCGTTAG	1283
<i>K2</i>	F: GACCCGATATTCATACTTGACAGAG R: CCTGAAGTAAATCGTAAATAGATGGC	641
<i>K5</i>	F: TGGTAGTGATGCTCGCGA R: CCTGAACCCACCCCAATC	280
<i>K20</i>	F: CGGTGCTACAGTGCATCATT R: GTTATACGATGCTCAGTCGC	741
<i>K54</i>	F: CATTAGCTCAGTGGTTGGCT R: GCTTGACAAACACCATACAG	881
<i>K57</i>	F: CTCAGGGCTAGAAGTGTCAT R: CACTAACCCAGAAAGTCGAG	1037
<i>fimH</i>	F: TGCTGCTGGGCTGGTCGATG R: CGGAGGGTGACGGTGACATC	909
<i>mrkD</i>	F: TTCTGCACAGCGGTCCC R: GATACCCGGCGTTTTCGTTAC	246
<i>magA</i>	F: GGTGCTCTTTACATCATTGC R: GCAATGGCCATTTGCGTTAG	1100
<i>rmpA</i>	F: ACTGGGCTACCTCTGCTTCA R: CTTGCATGAGCCATCTTTCA	516
<i>alls</i>	F: CCGAAACATTACGCACCTTT R: ATCACGAAGAGCCAGGTCAC	508
<i>wabG</i>	F: ACCATCGGCCATTTGATAGA R: CGGACTGGCAGATCCATATC	683
<i>uge</i>	F: TCTTCACGCCTTCCTTCACT R: GATCATCCGGTCTCCCTGTA	534
<i>aerobactin</i>	F: GCATAGGCGGATACGAACAT	556

	R: CACAGGGCAATTGCTTACCT	
<i>iroN</i>	F: GTCCGGCGGTAACTTCAGCC	829
	R: TCAGAATGAACTACCGCCC	
<i>iutA</i>	F: GGCTGGACATCATGGGAACTGG	300
	R: CGTCGGGAACGGGTAGAATCG	
<i>entB</i>	F: ATTCCTCAACTTCTGGGGC	371
	R: AGCATCGGTGGCGGTGGTCA	

Note: F represents the forward primer, and R represents the reverse primer.

Primers mentioned above for virulence-associated genes detection were designed from the reference (2).

Table S4 Detailed PCR reaction conditions for virulence-associated genes.

Name of genes	Annealing temperature (°C)	Extension time (s)
<i>K1</i>	55	70
<i>K2</i>	55	45
<i>K5</i>	55	35
<i>K20</i>	55	45
<i>K54</i>	55	55
<i>K57</i>	53	55
<i>aerobactin</i>	60	40
<i>rmpA</i>	60	40
<i>magA</i>	55	90
<i>iutA</i>	60	30
<i>uge</i>	55	40
<i>wabG</i>	56	30
<i>alls</i>	58	45
<i>iorN</i>	60	50
<i>wcaG</i>	53	30
<i>fimH</i>	55	55
<i>mrkD</i>	50	30
<i>entB</i>	52	30

Note: Crucial PCR conditions include the annealing temperature, and extension time for each virulence-associated gene were listed as above.

Supplementary references

1. R. R. Makharita, I. El-Kholy, H. F. Hetta, M. H. Abdelaziz, F. I. Hagagy, A. A. Ahmed and A. M. Algammal: Antibigram and Genetic Characterization of Carbapenem-Resistant Gram-Negative Pathogens Incriminated in Healthcare-Associated Infections. *Infect Drug Resist*, 13, 3991-4002 (2020) doi:10.2147/idr.S276975
2. E. D. Candan and N. Aksöz: Klebsiella pneumoniae: characteristics of carbapenem resistance and virulence factors. *Acta Biochim Pol*, 62(4), 867-74 (2015) doi:10.18388/abp.2015_1148