
Novel *ctx* Variants and Genotype-Phenotype Correlations among Clinical Isolates of Extended Spectrum Beta Lactamase-producing *Escherichia coli*

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Running Title: Genotype-phenotype correlations among ESBL *E. coli*

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Table S2: List of discs used for antimicrobial susceptibility testing

Antibiotic name	Abbreviation	Disc Content (μg)
Amoxicillin/ clavulanic acid	AMC	30
Ampicillin	AMP	10
Aztreonam	ATM	30
Cefalexin	LEX	30
Cefotaxime	CTX	30
Ceftazidime	CAZ	30
Cefuroxime	CXM	30
Cefoxitin	FOX	30
Imipenem	IPM	10
Penicillin G	P	10
Piperacillin	PRL	100
Piperacillin/ Tazobactam	TZP	110

Table S3: PCR primers for screening beta lactamase genes or determining the CTX genotypic group.

(Prod. = product; F = forward amplification primer; R = reverse amplification primer)

Target	Oligonucleotide sequence (5' to 3')	Prod. size	Purpose	Source
<i>bla</i> <i>TEM</i>	F: TCGTGTGCGCCCTTATTCCCTT R: GCGGTTAGCTCCTCCGGTCCT	409	PCR screening for beta-lactamase type	Ref 1
<i>bla</i> <i>SHV</i>	F: CGGCCCGCAGGATTGACT R: TCCCGGCGATTTGCTGATTC	391	PCR screening for beta-lactamase type	Ref 1
<i>bla</i> <i>CTX-M-1</i>	F: GTTAATTCGTCTCTCCAGAATAAGG R: GCTTATTCATCGCCACGTTATC	453	PCR screening for beta-lactamase type	This study
<i>bla</i> <i>CTX-M-2</i>	F: CGATGTGCAGTACCAGTAA R: TTAGTGACCAGAATCAGCGG	585	PCR screening for beta-lactamase type	Ref 2
<i>bla</i> <i>CTX-M-9</i>	F: GAGAGTGCAACGGATGATGT R: CGGTATTCAGCGTAGGTTCA	512	PCR screening for beta-lactamase type	This study
<i>bla</i> <i>CTX-M-15</i>	F: GCGCTACAGTACAGCGATAA R: TTTACCCAGCGTCAGATTCC	428	PCR screening for beta-lactamase type	This study
<i>bla</i> <i>CTX-M-25</i>	F: CTGGGTGTGGCGTTGATTA R: TCGTGCCATTGACGTGTT	213	PCR screening for beta-lactamase type	This study
Group 1 CTX	F: AAAAATCACTGCGCCAGTTC R: AGCTTATTCATCGCCACGTT	415	Multiplex PCR for CTX genotyping	Ref 3 cited in 4
Group 2 CTX	F: CGACGCTACCCCTGCTATT R: CCAGCGTCAGATTTTTCAGG	552	Multiplex PCR for CTX genotyping	Ref 3 cited in 4
Group 8 CTX	F: TCGCGTTAAGCGGATGATGC R: AACCCACGATGTGGGTAGC	666	Multiplex PCR for CTX genotyping	Ref 3 cited in 4
Group 9 CTX	F: CAAAGAGAGTGCAACGGATG R: ATTGGAAAGCGTTCATCACC	205	Multiplex PCR for CTX genotyping	Ref 3 cited in 4
Group 25 CTX	F: GCACGATGACATTCCGGG R: AACCCACGATGTGGGTAGC	327	Multiplex PCR for CTX genotyping	Ref 3 cited in 4
<i>ctx-M</i> Universal primers	MA1: SCSATGTGCAGYACCGATAA MA2: CCGCRATATGRTTGGTGGTG	variable	Amplifying all known <i>ctx</i> alleles	Ref 5

Figure S1.

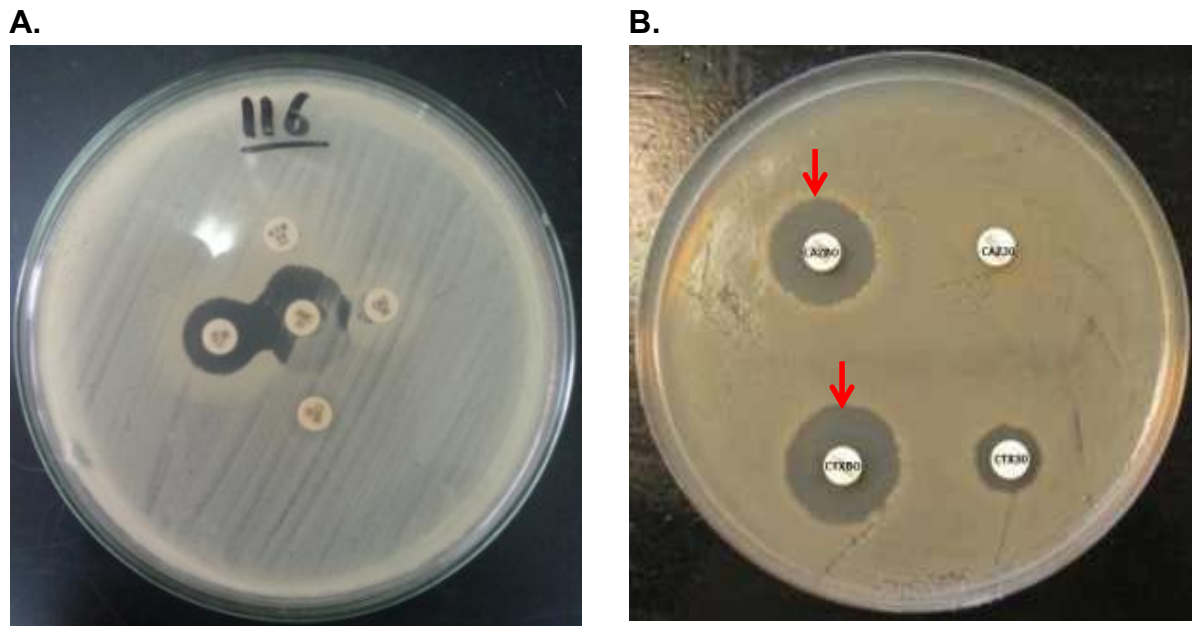


Figure S1. Representative *in vitro* tests used to determine the ESBL phenotypes

A. The typical keyhole pattern that indicates a positive ESBL isolate in the DDST. **B.** A representative positive result of the combined disc method: the zone diameter increased around the discs to which clavulanic acid was added (indicated with arrows)

FIGURE S2

A. BlastN best hit of *ctx-M-14.2*

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Query 1 CAGCTCGGTGGCCCGGGAGGCGTGACGGCTTTTGCCCGCGGATCGGCGATGAGACGTTT 60
      |||
Sbjct 386 CAGCTCGGTGGCCCGGGAGGCGTGACGGCTTTTGCCCGCGGATCGGCGATGAGACGTTT 327

Query 61 CGTCTGGATCGCACTGAACCTACGCTGAATACCGCCATTCCCGGCGACCCGAGAGACACC 120
      |||
Sbjct 326 CGTCTGGATCGCACTGAACCTACGCTGAATACCGCCATTCCCGGCGACCCGAGAGACACC 267

Query 121 ACCACGCCGCGGGCGATGGCACAGACGTTGCGTCAGCTTACGCTGGGTCATGCGCTGGGC 180
      |||
Sbjct 266 ACCACGCCGCGGGCGATGGCACAGACGTTGCGTCAGCTTACGCTGGGTCATGCGCTGGGC 207

Query 181 GAAACCCAGCGGGCGCAGTTGGTGACGTGGCTCAAAGGCAATACGACCGGCGCAGCCAGC 240
      |||
Sbjct 206 GAAACCCAGCGGGCGCAGTTGGTGACGTGGCTCAAAGGCAATACGACCGGCGCAGCCAGC 147

Query 241 ATTCTGGGCCGGCTTACCGACGTCGTGGACTGTAGGTGATAAGACCGGCAGCGCGACTAC 300
      |||
Sbjct 146 ATTCTGGGCCGGCTTACCGACGTCGTGGACTGTAGGTGATAAGACCGGCAGCGCGACTAC 87

Query 301 GGCACCACCAACCCATATCGCGG 322
      |||
Sbjct 86 GGCACCACCAATGATATGCGG 65

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B. BlastX best hit of *ctx-M-14.2*

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Query 1 QLGGPGGVTAFAARAIGDETFRLDRTEPTLNNTAIPGDPRDTTTPRAMAQTLRQLTLGHALG 180
      QLGGPGGVTAFAARAIGDETFRLDRTEPTLNNTAIPGDPRDTTTPRAMAQTLRQLTLGHALG
Sbjct 72 QLGGPGGVTAFAARAIGDETFRLDRTEPTLNNTAIPGDPRDTTTPRAMAQTLRQLTLGHALG 131

Query 181 ETQRAQLVTWLKGNNTGAASIRAGLPTSWTVGDKTGSGDYGTTNHIA 321
      ETQRAQLVTWLKGNNTGAASIRAGLPTSWTVGDKTGSGDYGTTN IA
Sbjct 132 ETQRAQLVTWLKGNNTGAASIRAGLPTSWTVGDKTGSGDYGTTNIIA 178

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C. BlastN best hit of *ctx-M-15.2*

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Query      1  GCGGTGCTGCTGAGAAAGTGAAGCGAACCGAATCTGTTAAATCAGCGAGTTGAGATCAAA  60
          |||
Sbjct    2593 GCGGTGCTGAAAGAAAGTGAAGCGAACCGAATCTGTTAAATCAGCGAGTTGAGATCAAA  2652

Query     61  AAATCTGACCTTGTTAACTATAATCCGATTGCGGAAAAGCACGTCAATGGGACGATGTCA  120
          |||
Sbjct    2653 AAATCTGACCTTGTTAACTATAATCCGATTGCGGAAAAGCACGTCAATGGGACGATGTCA  2712

Query     121  CTGGCTGAGCTTAGCGCGGCCGCTACAGTACAGCGATAACGTGGCGATGAATAAGCTG  180
          |||
Sbjct    2713 CTGGCTGAGCTTAGCGCGGCCGCTACAGTACAGCGATAACGTGGCGATGAATAAGCTG  2772

Query     181  ATTGCTCACGTTGGCGGCCCGCTAGCGTCACCGCGTTCGCCCACAGCTGGGAGACGAA  240
          |||
Sbjct    2773 ATTGCTCACGTTGGCGGCCCGCTAGCGTCACCGCGTTCGCCCACAGCTGGGAGACGAA  2832

Query     241  ACGTTCGCTCTCGACCGTACCGAGCCGACGTAAACACCGCCATTCCGGGCGATCCGCGT  300
          |||
Sbjct    2833 ACGTTCGCTCTCGACCGTACCGAGCCGACGTAAACACCGCCATTCCGGGCGATCCGCGT  2892

Query     301  GATACCACTTCACCTCGGGCAATGGCGCAAACCTGCGGAATCTGACGCTGGGTAAAGCA  360
          |||
Sbjct    2893 GATACCACTTCACCTCGGGCAATGGCGCAAACCTGCGGAATCTGACGCTGGGTAAAGCA  2952

Query     361  TTGGGCGACAGCCAACGGGCGCAGCTGGTGACATGGATGAAAGGCAATACCACCGGTGCA  420
          |||
Sbjct    2953 TTGGGCGACAGCCAACGGGCGCAGCTGGTGACATGGATGAAAGGCAATACCACCGGTGCA  3012

Query     421  GCGAGCATTACAGCTGGACTGCCTGCTTCTGGGTTGTGGGGATAAAACCGGCAGCGGT  480
          |||
Sbjct    3013 GCGAGCATTACAGCTGGACTGCCTGCTTCTGGGTTGTGGGGATAAAACCGGCAGCGGT  3072

Query     481  GGCTATGGCACCACCAAC  498
          |||
Sbjct    3073 GGCTATGGCACCACCAAC  3090

```

D. BlastX best hit of *ctx-M-15.2*

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Query      1  AVLRSSESEPILLNQRVEIKKSDLVNYNPIAEKHVNGTMSLAELSAAALQYSDNVAMNKL  180
          |
Sbjct     12  AVLKSESEPILLNQRVEIKKSDLVNYNPIAEKHVNGTMSLAELSAAALQYSDNVAMNKL  71

Query     181  IAHVGGPASVTAFARQLGDETFRLDRTEPTLNIAIPGDPRDTTSPRAMAQLRNLTLGKA  360
          |||
Sbjct     72  IAHVGGPASVTAFARQLGDETFRLDRTEPTLNIAIPGDPRDTTSPRAMAQLRNLTLGKA  131

Query     361  LGDSQRAQLVTWMKGNTTGAASIQAGLPASWVVGDKTGSGGYGTTN  498
          |||
Sbjct    132  LGDSQRAQLVTWMKGNTTGAASIQAGLPASWVVGDKTGSGGYGTTN  177

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Figure S2: BlastN and BlastX alignment of the newly identified *ctx* alleles to their best matching genes (variant nucleotides or amino acids are colored in red)

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