

**Structure and tailspike glycosidase machinery of ORF212 from *E. coli* O157:H7 phage CBA120 (TSP3)**

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## Supplementary Material

**Table S1: Bacterial strains used in this study**

<b>Organism</b>	<b>Strain</b>	<b>Source</b>	<b>Notes</b>
<i>Escherichia coli</i>	Rosetta-gami	Novagen	
<i>Escherichia coli</i>	700728	ATCC	O157:H7 Non-toxigenic strain
<i>Escherichia coli</i>	43894	ATCC	O157:H7 Toxigenic strain
<i>Escherichia coli</i>	35218	ATCC	
<i>Escherichia coli</i>	TEA023	Ref. 11	O157:H7 knockout mutant EDL933 $\Delta$ galU::aad-7
<i>Escherichia coli</i>	TEA026	Ref. 11	O157:H7 knockout mutant EDL933 $\Delta$ galETKM::aad-7
<i>Klebsiella pneumoniae</i>	700603	ATCC	
<i>Pseudomonas aeruginosa</i>	27853	ATCC	
<i>Acinetobacter baumannii</i>	BAA-1605	ATCC	

**Table S2: Plasmids used in this study**

<b>Plasmid</b>	<b>Relevant properties</b>	<b>Source or Reference</b>
pBAD24	Cloning and expression containing the arabinose pBAD promoter	Qiagen
pBAD24:: <i>TSP3</i>	WT <i>TSP3</i>	This work
pBAD24:: <i>TSP3</i> <sub>E362Q</sub>	Site-direct mutagenesis of the 362 <sup>nd</sup> amino acid, glutamic acid to glutamine	This work
pBAD24:: <i>TSP3</i> <sub>D383N</sub>	Site-direct mutagenesis of the 383 <sup>rd</sup> amino acid, aspartic acid to asparagine	This work
pBAD24:: <i>TSP3</i> <sub>D426N</sub>	Site-direct mutagenesis of the 426 <sup>th</sup> amino acid, aspartic acid to asparagine	This work
pBAD24:: <i>TSP3</i> <sub>E362Q D383N</sub>	Site-direct mutagenesis of the 362 <sup>nd</sup> amino acid, glutamic acid to glutamine, and 383 <sup>rd</sup> amino, acid aspartic acid to asparagine	This work
pBAD24:: <i>TSP3</i> <sub>E362Q D426N</sub>	Site-direct mutagenesis of the 362 <sup>nd</sup> amino acid, glutamic acid to glutamine, and 426 <sup>th</sup> amino acid, aspartic acid to asparagine	This work
pBAD24:: <i>TSP3</i> <sub>D383Q D426N</sub>	Site-direct mutagenesis of the 383 <sup>rd</sup> amino acid, aspartic acid to asparagine, and 426 <sup>th</sup> amino acid, aspartic acid to asparagine	This work
pBAD24:: <i>TSP3</i> <sub>E362Q D383N D426N</sub>	Site-direct mutagenesis of the 362 <sup>nd</sup> amino acid, glutamic acid to glutamine, 383 <sup>rd</sup> amino acid, aspartic acid to asparagine, and 426 <sup>th</sup> amino acid, aspartic acid to asparagine	This work

**Table S3: Primers used in this study.**

Purpose	Template	Sequence (5' > 3')*
Site-directed mutagenesis E326Q	pBAD24:: <i>TSP3</i>	[Phos]-ATTCAGTTTATTAGCCAG <b>G</b> AGCCCGAAATTTAC
Site-directed mutagenesis D383N	pBAD24:: <i>TSP3</i>	[Phos]-GTTTGGGATACCGCAA <b>A</b> CCTGGCAATTACCAA
Site-directed mutagenesis D426N	pBAD24:: <i>TSP3</i>	[Phos]- ACCTGTAGCGTTTTTA <b>A</b> CGCCGTTTCGTGGTCAT
Site-directed mutagenesis E326Q D383N	pBAD24:: <i>TSP3<sub>E362Q</sub></i>	[Phos]- GTTTGGGATACCGCAA <b>A</b> CCTGGCAATTACCAA
Site-directed mutagenesis E326Q D426N	pBAD24:: <i>TSP3<sub>E362Q</sub></i>	[Phos]- ACCTGTAGCGTTTTTA <b>A</b> CGCCGTTTCGTGGTCAT
Site-directed mutagenesis E326Q D383N D426N	pBAD24:: <i>TSP3<sub>E362Q</sub></i> <i>D383N</i>	[Phos]- ACCTGTAGCGTTTTTA <b>A</b> CGCCGTTTCGTGGTCAT

\* Point mutations bold