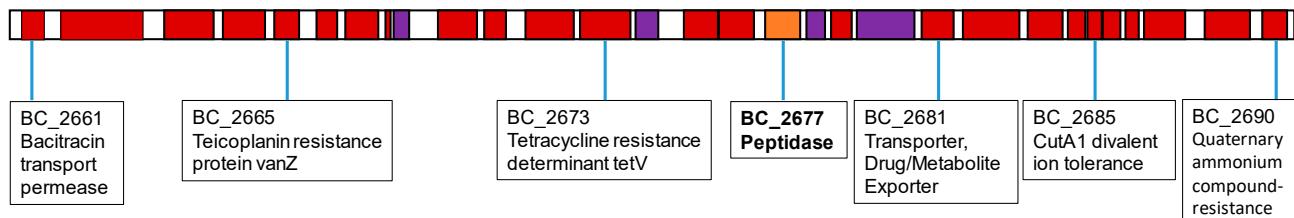


## Supplemental Information

### Characterization of LysBC17, a lytic endopeptidase from *Bacillus cereus*

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**Supplemental Figure 1.** Drug and metal resistance genes near the BC\_2677 locus in the *B. cereus* ATCC 14579 genome (GenBank: AE016877.1). The 32 kb window displayed covers nucleotides 2643000 – 2675000. No holin gene or other phage genes are nearby. The BC\_2677 gene does not encode a signal peptide. This suggests BC\_2677 (LysBC17) is an autolysin. Orange and red boxes are coding regions in the same orientation, and purple boxes are coded on the complementary DNA strand.

PlyBC17	1	MKYHNRN <span style="background-color: black; color: black;">V</span> SNL <span style="background-color: black; color: black;">I</span> NK <span style="background-color: black; color: black;">K</span> LADNT <span style="background-color: black; color: black;">T</span> <span style="background-color: black; color: black;">K</span> AAAFK <span style="background-color: black; color: black;">W</span> YQYC <span style="background-color: black; color: black;">I</span> D <span style="background-color: black; color: black;">D</span> NGIEVL <span style="background-color: black; color: black;">I</span> YET <span style="background-color: black; color: black;">T</span> RT <span style="background-color: black; color: black;">V</span> EQQRE <span style="background-color: black; color: black;">Y</span> VRKGAS <span style="background-color: black; color: black;">T</span> QM
Wip1	1	MYHNRN <span style="background-color: black; color: black;">I</span> ANLE <span style="background-color: black; color: black;">K</span> LAPHT <span style="background-color: black; color: black;">P</span> QKA <span style="background-color: black; color: black;">K</span> WYQYC <span style="background-color: black; color: black;">V</span> ENGIEVL <span style="background-color: black; color: black;">I</span> YET <span style="background-color: black; color: black;">T</span> RT <span style="background-color: black; color: black;">I</span> EQQRE <span style="background-color: black; color: black;">N</span> VRKGAS <span style="background-color: black; color: black;">T</span> QM

PlyBC17	61	RSYHLVGQALDFVP <span style="background-color: black; color: black;">I</span> QS <span style="background-color: black; color: black;">N</span> G <span style="background-color: black; color: black;">C</span> TED <span style="background-color: black; color: black;">W</span> NG <span style="background-color: black; color: black;">Y</span> N <span style="background-color: black; color: black;">K</span> EPWASAIR <span style="background-color: black; color: black;">Y</span> AK <span style="background-color: black; color: black;">Q</span> I <span style="background-color: black; color: black;">G</span> F <span style="background-color: black; color: black;">E</span> WGGDW <span style="background-color: black; color: black;">K</span> GF <span style="background-color: black; color: black;">F</span> V <span style="background-color: black; color: black;">D</span> S <span style="background-color: black; color: black;">P</span> <span style="background-color: black; color: black;">H</span> L <span style="background-color: black; color: black;">Q</span> Y	120
Wip1	61	KSYHLVGQALDFVP <span style="background-color: black; color: black;">A</span> RE <span style="background-color: black; color: black;">-E</span> EV <span style="background-color: black; color: black;">Y</span> WD <span style="background-color: black; color: black;">G</span> YY <span style="background-color: black; color: black;">R</span> ND <span style="background-color: black; color: black;">I</span> Q <span style="background-color: black; color: black;">K</span> A <span style="background-color: black; color: black;">I</span> G <span style="background-color: black; color: black;">Y</span> AK <span style="background-color: black; color: black;">S</span> I <span style="background-color: black; color: black;">G</span> F <span style="background-color: black; color: black;">E</span> WGGDW <span style="background-color: black; color: black;">K</span> GF <span style="background-color: black; color: black;">F</span> V <span style="background-color: black; color: black;">D</span> S <span style="background-color: black; color: black;">P</span> X <span style="background-color: black; color: black;">L</span> Q <span style="background-color: black; color: black;">Y</span>	119

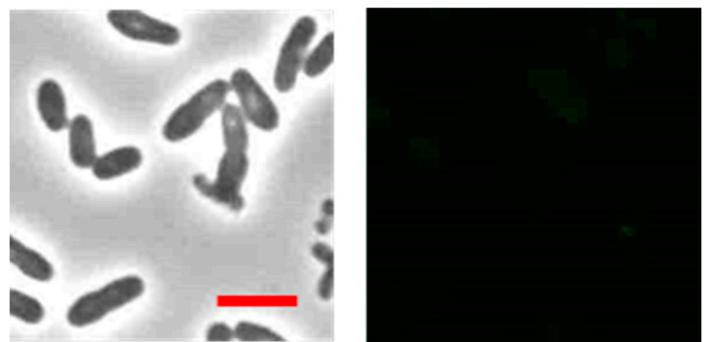
**Supplemental Figure 3.** Clustal Omega alignment and BoxShade view of the LysBC17 catalytic domain (residues 1-120) and the *Bacillus* phage Wip1 endolysin catalytic domain (YP\_008433324, residues 1-119). Black boxes represent identity and grey boxes represent similarity.

PlyBC17	131	G <span style="background-color: black; color: black;">K</span> G <span style="background-color: black; color: black;">A</span> -QNVVTP <span style="background-color: black; color: black;">P</span> PT <span style="background-color: black; color: black;">S</span> ND <span style="background-color: black; color: black;">N</span> V <span style="background-color: black; color: black;">G</span> VAY <span style="background-color: black; color: black;">I</span> NG <span style="background-color: black; color: black;">S</span> SV <span style="background-color: black; color: black;">N</span> V <span style="background-color: black; color: black;">L</span> R <span style="background-color: black; color: black;">K</span> G <span style="background-color: black; color: black;">P</span> GT <span style="background-color: black; color: black;">G</span> Y <span style="background-color: black; color: black;">Y</span> VIR <span style="background-color: black; color: black;">Q</span> LG <span style="background-color: black; color: black;">K</span> G <span style="background-color: black; color: black;">E</span> SY <span style="background-color: black; color: black;">K</span> V <span style="background-color: black; color: black;">V</span> G <span style="background-color: black; color: black;">Q</span> T <span style="background-color: black; color: black;">I</span> N <span style="background-color: black; color: black;">S</span> WL
Waukesha92	160	GG <span style="background-color: black; color: black;">G</span> S <span style="background-color: black; color: black;">P</span> V <span style="background-color: black; color: black;">M</span> P <span style="background-color: black; color: black;">T</span> P <span style="background-color: black; color: black;">I</span> PP <span style="background-color: black; color: black;">S</span> ND <span style="background-color: black; color: black;">G</span> T <span style="background-color: black; color: black;">K</span> VAY <span style="background-color: black; color: black;">I</span> NG <span style="background-color: black; color: black;">D</span> SV <span style="background-color: black; color: black;">N</span> V <span style="background-color: black; color: black;">L</span> R <span style="background-color: black; color: black;">K</span> G <span style="background-color: black; color: black;">P</span> GT <span style="background-color: black; color: black;">G</span> Y <span style="background-color: black; color: black;">Y</span> VIR <span style="background-color: black; color: black;">K</span> LG <span style="background-color: black; color: black;">K</span> G <span style="background-color: black; color: black;">E</span> CY <span style="background-color: black; color: black;">Q</span> V <span style="background-color: black; color: black;">V</span> GE <span style="background-color: black; color: black;">S</span> NG <span style="background-color: black; color: black;">W</span> L

PlyBC17	190	N <span style="background-color: black; color: black;">L</span> GG <span style="background-color: black; color: black;">D</span> QW <span style="background-color: black; color: black;">V</span> YND <span style="background-color: black; color: black;">P</span> SY <span style="background-color: black; color: black;">I</span> RY <span style="background-color: black; color: black;">T</span> G <span style="background-color: black; color: black;">G</span> GN <span style="background-color: black; color: black;">V</span> P <span style="background-color: black; color: black;">A</span> T <span style="background-color: black; color: black;">S</span> Q <span style="background-color: black; color: black;">S</span> S <span style="background-color: black; color: black;">N</span> D <span style="background-color: black; color: black;">G</span> V <span style="background-color: black; color: black;">V</span> V <span style="background-color: black; color: black;">T</span> I <span style="background-color: black; color: black;">I</span> AD <span style="background-color: black; color: black;">V</span> L <span style="background-color: black; color: black;">R</span> V <span style="background-color: black; color: black;">T</span> G <span style="background-color: black; color: black;">P</span> GT <span style="background-color: black; color: black;">N</span> Y <span style="background-color: black; color: black;">G</span> IV <span style="background-color: black; color: black;">V</span> KN <span style="background-color: black; color: black;">V</span> Y
Waukesha92	220	N <span style="background-color: black; color: black;">L</span> GG <span style="background-color: black; color: black;">D</span> QW <span style="background-color: black; color: black;">V</span> YND <span style="background-color: black; color: black;">S</span> SY <span style="background-color: black; color: black;">I</span> RY <span style="background-color: black; color: black;">T</span> G <span style="background-color: black; color: black;">E</span> N <span style="background-color: black; color: black;">A</span> P <span style="background-color: black; color: black;">A</span> P <span style="background-color: black; color: black;">S</span> K <span style="background-color: black; color: black;">P</span> S <span style="background-color: black; color: black;">N</span> D <span style="background-color: black; color: black;">G</span> I <span style="background-color: black; color: black;">V</span> V <span style="background-color: black; color: black;">T</span> I <span style="background-color: black; color: black;">T</span> AD <span style="background-color: black; color: black;">V</span> L <span style="background-color: black; color: black;">R</span> V <span style="background-color: black; color: black;">T</span> G <span style="background-color: black; color: black;">P</span> GT <span style="background-color: black; color: black;">N</span> Y <span style="background-color: black; color: black;">G</span> IV <span style="background-color: black; color: black;">V</span> KN <span style="background-color: black; color: black;">V</span> Y

PlyBC17	250	Q <span style="background-color: black; color: black;">G</span> E <span style="background-color: black; color: black;">K</span> Y <span style="background-color: black; color: black;">Q</span> S <span style="background-color: black; color: black;">F</span> GY <span style="background-color: black; color: black;">K</span> D <span style="background-color: black; color: black;">G</span> W <span style="background-color: black; color: black;">Y</span> N <span style="background-color: black; color: black;">V</span> GG <span style="background-color: black; color: black;">G</span> N <span style="background-color: black; color: black;">Q</span> W <span style="background-color: black; color: black;">V</span> S <span style="background-color: black; color: black;">G</span> E <span style="background-color: black; color: black;">Y</span> V <span style="background-color: black; color: black;">T</span> F <span style="background-color: black; color: black;">V</span> K <span style="background-color: black; color: black;">K</span>	281
Waukesha92	280	Q <span style="background-color: black; color: black;">S</span> E <span style="background-color: black; color: black;">R</span> Y <span style="background-color: black; color: black;">Q</span> S <span style="background-color: black; color: black;">W</span> GY <span style="background-color: black; color: black;">R</span> D <span style="background-color: black; color: black;">G</span> W <span style="background-color: black; color: black;">Y</span> N <span style="background-color: black; color: black;">V</span> GG <span style="background-color: black; color: black;">G</span> D <span style="background-color: black; color: black;">Q</span> W <span style="background-color: black; color: black;">V</span> S <span style="background-color: black; color: black;">G</span> E <span style="background-color: black; color: black;">Y</span> V <span style="background-color: black; color: black;">K</span> E <span style="background-color: black; color: black;">E</span> K <span style="background-color: black; color: black;">K</span>	311

**Supplemental Figure 2.** Clustal Omega alignment and BoxShade view of the LysBC17 binding domain (residues 231-281) and the *Bacillus* phage Waukesha92 endolysin binding domain (YP\_009099314, residues 160-311). Black boxes represent identity and grey boxes represent similarity.



**Supplemental Figure 4.** LysBC17 GFP-CWB binding to *B. cereus* ATCC 13061. Bacterial cells and GFP-labeled LysBC17 CWB were mixed together, washed, and viewed by microscopy per the Methods. 1000X phase-contrast image (left panel) is shown with its corresponding fluorescent images (right panel). Exposure time for the fluorescent image was 100 ms, identical to the time used in Figure 5. Scale bar = 5  $\mu$ m.