

## Supplementary Material (SM)

**Table S1. Spectrophotometer reading (OD<sub>600nm</sub>) of *B/1821L* culture after treatment with the mitomycin C at various concentrations.**

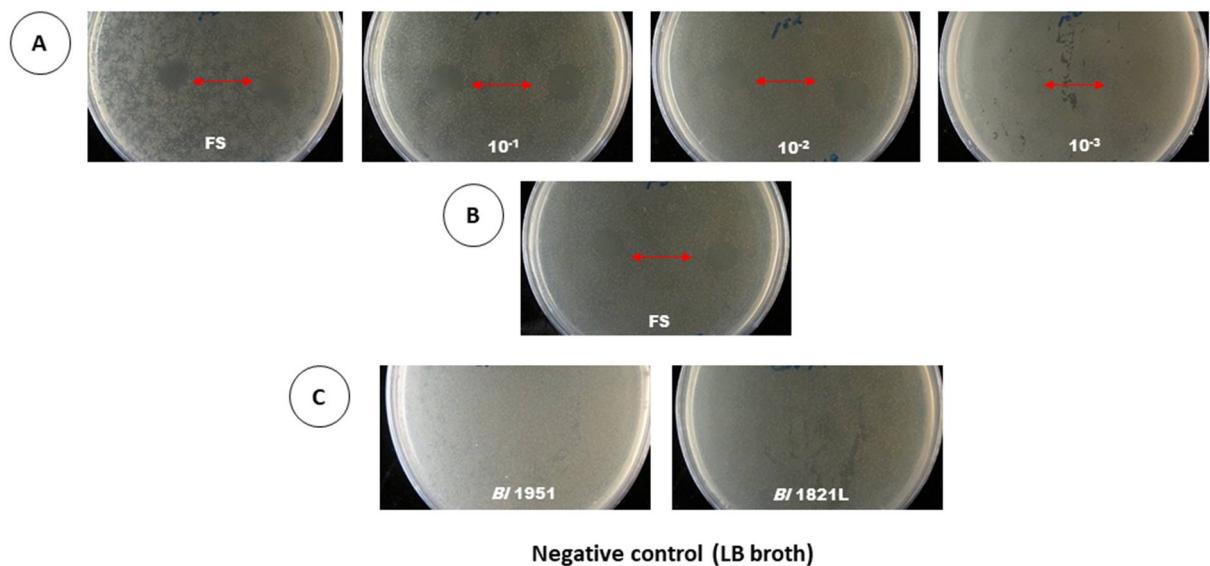
| Treatments                        | Time intervals<br>(Hours) |      |      |      |      | % Decrease<br>in OD <sub>600nm</sub> |
|-----------------------------------|---------------------------|------|------|------|------|--------------------------------------|
|                                   | 0                         | 2    | 4    | 6    | 24   |                                      |
| Mitomycin C<br>(1 µg/mL)          | 1.41                      | 1.44 | 1.46 | 1.48 | 0.71 | 49.65                                |
| Mitomycin C<br>(3 µg/mL)          | 1.56                      | 1.56 | 1.57 | 1.58 | 0.92 | 41.03                                |
| Control                           | 1.30                      | 1.34 | 1.47 | 1.49 | 1.35 | -3.85                                |
| *LSD (5%)<br>(Control vs Treated) | 0.18                      | 0.15 | 0.12 | 0.12 | 0.37 | 24.49                                |
| LSD (5%)<br>(**MMC1 vs ***MMC3)   | 0.16                      | 0.14 | 0.12 | 0.11 | 0.34 | 22.68                                |

Note: \*LSD= Least significant digit

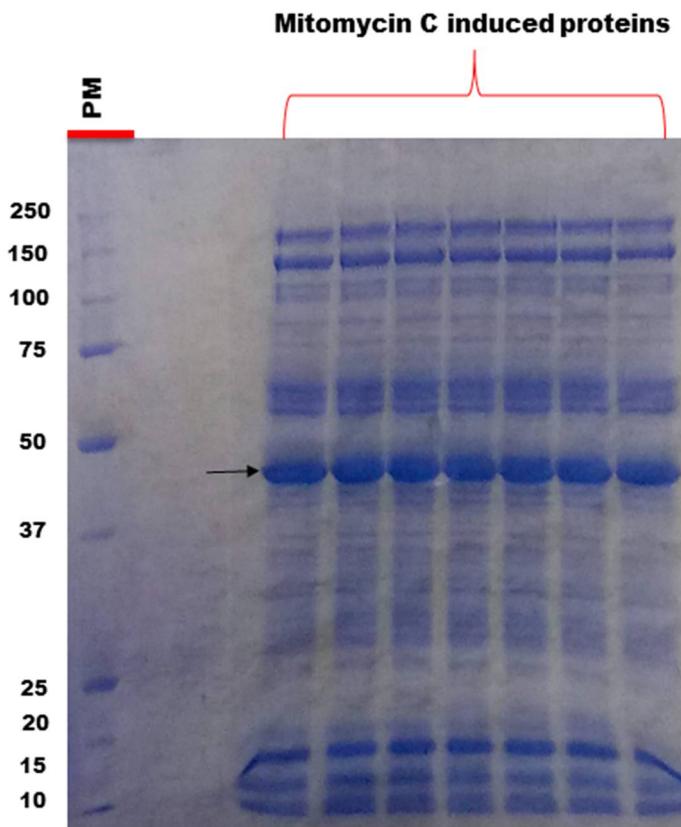
\*\*MMC1= Mitomycin C @ 1 µg/mL

\*\*\*MMC3= Mitomycin C @ 3 µg/mL

*Brevibacillus laterosporus* 1821L cell free supernatant activity after PEG 8000 precipitation



**Figure S1. Antibacterial activity of *B/l* 1821L cell free supernatant after PEG 8000 precipitation in the serial dilutions assay test against *B/l* 1951 as the host bacterium (A) and *B/l* 1821L as the host bacterium (B). Arrows (red colour) denote the zones of inhibition due to the activity of PEG 8000 precipitated putative antibacterial proteins. LB broth was used as a negative control against both the strains (*B/l* 1951 & *B/l* 1821L) (C).**

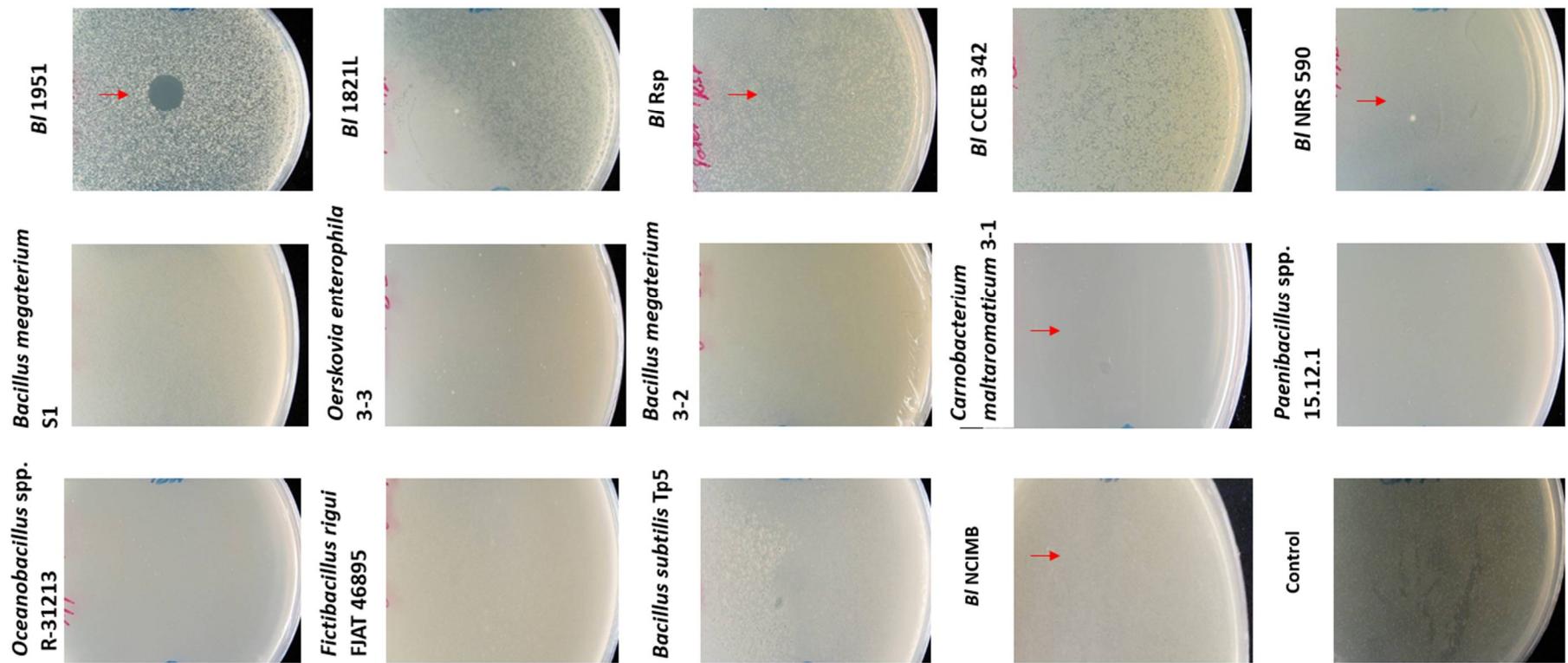


**Figure S2. SDS-PAGE of mitomycin C induced culture of *B/ 1821L* showing a prominent band of ~48 kD (shown with dark arrow).**

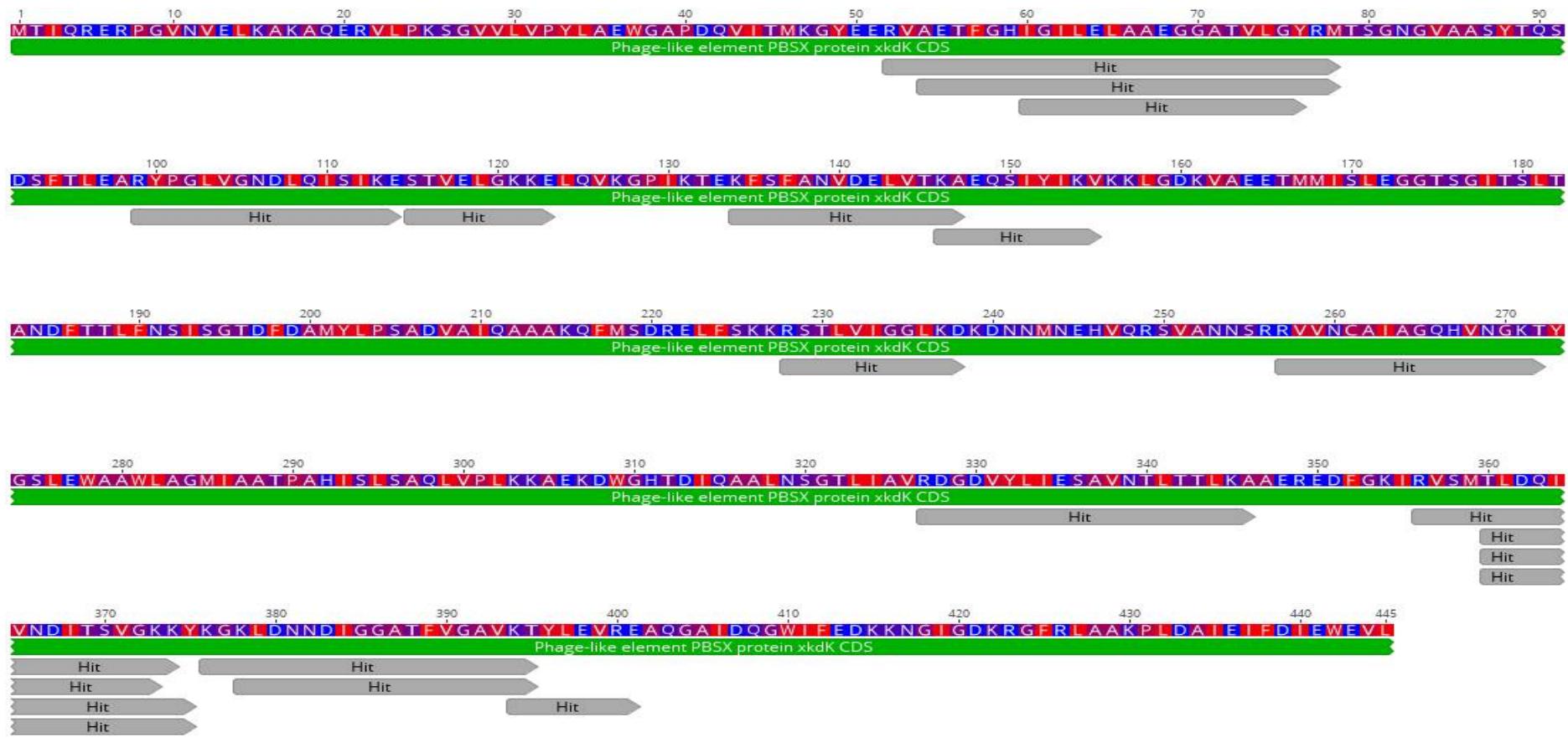
Note: Based on its prominence, ~48 kD protein band was hypothesised to be involved in the putative antibacterial activity. Therefore, the band was excised for preliminary N-terminal sequencing and the resultant short sequence amino acids revealed several hits (covering 70% of the amino acid sequence) to the loci A0A518VEB0 in the *B/ 1821L* genome (NZ\_CP033464.1) encoding a predicted defective phage protein, similar to that encoded by the *Bs* 168 phage-like element PBSX gene *xkdK*. Furthermore, these findings were authenticated with the subsequent TEM examination, SDS-PAGE, N-terminal sequencing, and bioinformatic analysis of purified ~48 kD protein.

Refer to N-terminal sequencing of ~48 kD crude protein:  
<https://data.lincoln.ac.nz/n downloader/files/30948223>

*Brevibacillus laterosporus* 1821L induced cultures after PEG 8000 precipitation



**Figure S3.** Antibacterial activity of *BI* 1821L induced culture cell free supernatant after PEG 8000 precipitation against various gram-positive bacteria. Arrows (red colour) denote the zone of inhibition due to the activity of PEG 8000 precipitated putative antibacterial proteins.

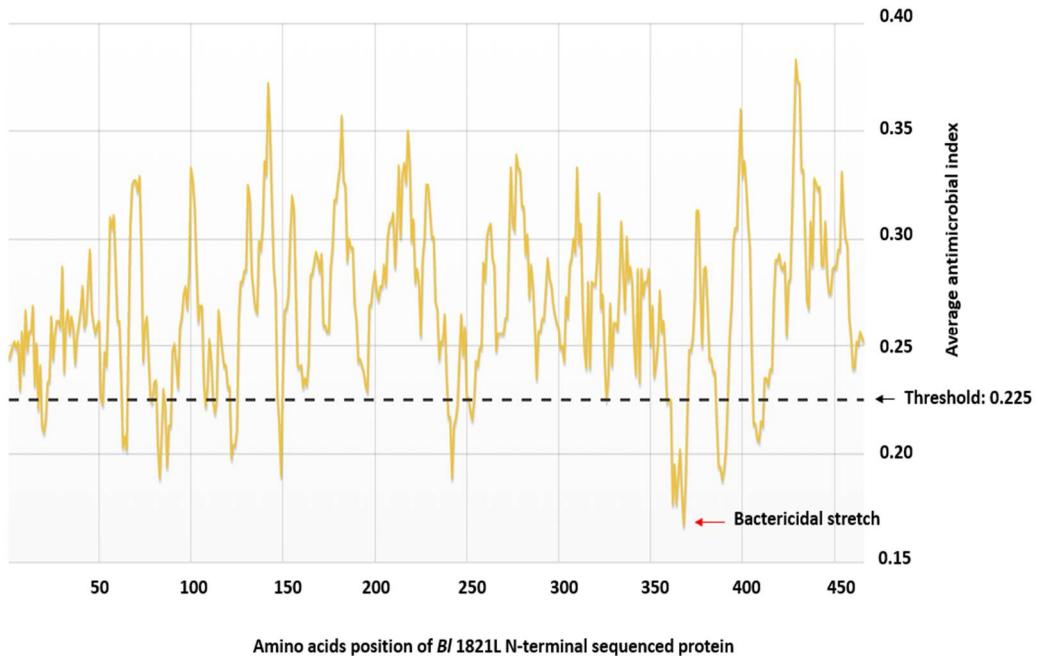


**Figure S4.** N-terminal sequenced short amino acid match hits to the putative phage tail protein (~48 kD) encoded in *B1* 1821L genome.

**Refer to N-terminal sequencing of ~48 kD purified protein: (<https://data.lincoln.ac.nz/nDownloader/files/30948616>)**

**Table S2. Similar proteins to the ~48 kD identified putative phage tail protein of BI 1821L in the Uniprot database.**

| Similar proteins          | Organism  | Gene name                                   | Length |
|---------------------------|---|---|--------|
| Phage tail protein        | <i>Brevibacillus laterosporus</i><br>( <i>Bacillus laterosporus</i> ) | C4A76_07870,<br>C4A77_13935,<br>D5F52_00915 | 445    |
| Phage tail protein        | <i>Brevibacillus laterosporus</i><br>( <i>Bacillus laterosporus</i> ) | EX87_06735                                  | 445    |
| Phage tail-sheath protein | <i>Brevibacillus laterosporus</i> LMG 15441                           | BRLA_c036460                                | 462    |
| Uncharacterised protein   | <i>Brevibacillus borstelensis</i> GI-9                                | BLGI_826                                    | 445    |
| Phage tail protein        | <i>Brevibacillus laterosporus</i><br>( <i>Bacillus laterosporus</i> ) | EEL32_11960                                 | 445    |
| Phage tail protein        | <i>Brevibacillus laterosporus</i> SKDU 10                             | AYJO8_14030                                 | 445    |
| Phage tail protein        | <i>Brevibacillus laterosporus</i><br>( <i>Bacillus laterosporus</i> ) | EX87_02320                                  | 445    |
| Phage tail protein        | <i>Brevibacillus laterosporus</i><br>( <i>Bacillus laterosporus</i> ) | D5F52_16160                                 | 445    |
| Phage tail protein        | <i>Brevibacillus laterosporus</i><br>( <i>Bacillus laterosporus</i> ) | C4A76_21720                                 | 445    |



**Figure S5. AMPA analysis of identified phage tail-sheath protein of BI 1821L. Identified bactericidal motif is indicated is pointed with red arrow.**

**1** MNGGTFTTGKEKERAGIYFNFKTTAQERVSLSERGTVALPVASSWGEAKTFVSISSVEDL  
**61** NKKVGLSIDDPSSLRLREAKKNAKTVLMLYRLTEGVRSADIAEGVKATAVYGGTKGNDII  
**121** IRINQNVLDA NSFDVTTYMDESEVDKQTVKKAELTANGYVTFTGTGDLSSIPLTGSEG  
**181** DTAAE TLNASAGIRLSGGTDKAPVNSDYTDLAAETESFDVIALPVAEGDQLKATFAAF  
**241** IKRLRDGQQKVQGV TANYAGDYEGIINVTEGVLL EDGTEVTPDKATAWVAGASAGATN  
**301** QSLTFVEYEGA DVVLHRLDHDTIVERLGKG EFLFTDARDKS VSKEDINS LVTFTAEN  
**361** KKFAKNKIVRVLD AVNNNDLTRELKALIKSRKGSGSDIPASEDGLQYVKTMITQYMTTLQD  
**421** AGGITGFDSDEDITISMNEDRDGFLIDLAVQPV DAAEKFYFNVEVN

**Figure S6. AMPA analysis of identified phage-like element PBSX protein XkdK of BI 1821L. Amino acids (360-373) corresponding to the bactericidal stretch are highlighted in red colour.**

**Table S3.** Effect of crude *Bt* 1821L putative antibacterial proteins (ABPs) on the number of viable cells of *Bt* 1821L and *Bt* 1951 after incubation at 30°C for various time intervals. Data presents the mean values of four experiments. Values of % decrease/increase in the number of viable cells are calculated from CFUs values of corresponding time intervals.

| Time intervals<br>(Hours) | <i>Bt</i> 1821L       | <i>Bt</i> 1821L<br>+<br><i>Bt</i> 1821L crude ABPs | % Decrease/increase<br>in no. of viable cells | <i>Bt</i> 1951      | <i>Bt</i> 1951<br>+<br><i>Bt</i> 1821L crude ABPs | % Decrease/increase<br>in no. of viable cells | *LSD (5%) |
|---------------------------|-----------------------|--|---|---------------------|---|---|-----------|
| 1                         | 4.46E+06<br>(6.650)** | 5.56E+06<br>(6.745)                                | -24.65  | 3.18E+06<br>(6.502) | 4.19E+06<br>(6.622)                               | -31.89  | 0.350     |
| 3                         | 3.70E+06<br>(6.568)   | 3.85E+06<br>(6.585)                                | -4.05   | 3.64E+06<br>(6.561) | 4.93E+06<br>(6.692)                               | -35.40  | 0.281     |
| 6                         | 3.58E+06<br>(6.553)   | 2.50E+06<br>(6.398)                                | 30.07   | 3.74E+06<br>(6.573) | 5.80E+06<br>(6.763)                               | -55.18  | 0.377     |
| 12                        | 1.18E+07<br>(7.071)   | 1.10E+07<br>(7.043)                                | 6.36  | 9.15E+06<br>(6.961) | 1.33E+07<br>(7.123)                               | -45.22  | 0.312     |
| 18                        | 1.35E+07<br>(7.131)   | 1.67E+07<br>(7.223)                                | -23.57  | 1.05E+07<br>(7.023) | 1.17E+07<br>(7.068)                               | -11.03  | 0.228     |
| 24                        | 1.72E+07<br>(7.236)   | 1.50E+07<br>(7.177)                                | 12.71   | 5.58E+06<br>(6.746) | 7.89E+06<br>(6.897)                               | -41.48  | 0.317     |

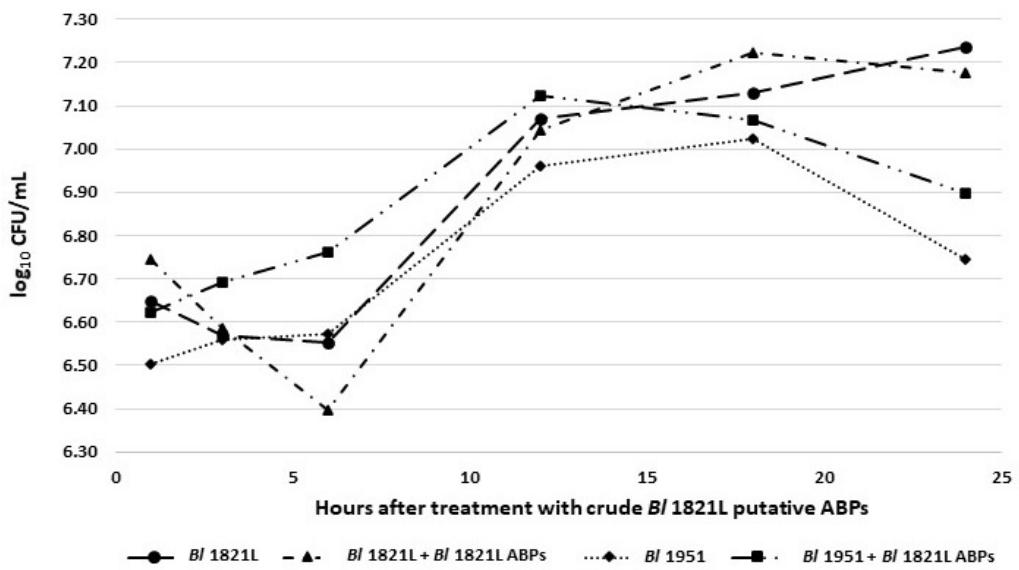
\*=Least significant difference

\*\*=The values in parenthesis indicate the converted value of number of viable cells (CFU/mL) into log<sub>10</sub> CFU/mL.

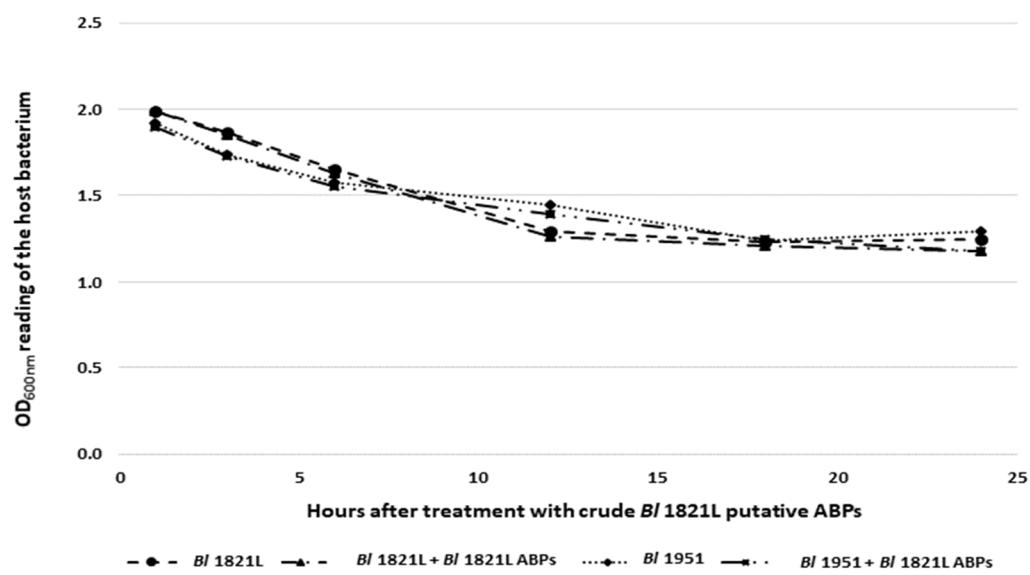
**Table S4.** Effect of crude *B.1821* putative antibacterial proteins (ABPs) on the OD<sub>600nm</sub> reading of *B.1821L* and *B.1951* after incubation at 30°C for various time intervals. Data presents the mean values of four experiments.

| Time intervals<br>(Hours)   | <i>B.1821L</i> | <i>B.1821L</i><br>+<br><i>B.1821L</i> crude ABPs | % Decrease/increase<br>in OD <sub>600nm</sub> reading | <i>B.1951</i> | <i>B.1951</i><br>+<br><i>B.1821L</i> crude ABPs | % Decrease/increase<br>in OD <sub>600nm</sub> reading | *LSD (5%) |
|---|----------------|--|---|---------------|---|---|-----------|
| 1   | 1.99           | 1.99   | 0.00  | 1.92          | 1.90  | 1.14  | 0.061     |
| 3   | 1.87           | 1.85   | 0.67  | 1.74          | 1.73  | 0.43  | 0.121     |
| 6   | 1.65           | 1.63   | 1.21  | 1.58          | 1.55  | 1.51  | 0.148     |
| 12  | 1.29           | 1.26   | 2.32  | 1.45          | 1.39  | 4.57  | 0.135     |
| 18  | 1.23           | 1.21   | 1.63  | 1.24          | 1.25  | -0.10   | 0.227     |
| 24  | 1.25           | 1.18   | 5.51  | 1.29          | 1.18  | 8.36  | 0.237     |
| <b>% Decrease from<br/>the start (1 hour) to<br/>the end (24 hours)<br/>of incubation</b> | 37.2%          | 40.7%  |   | 32.8%         | 37.9%   |   |           |

\*=Least significant difference



**Figure S7.** Number of viable cells ( $\log_{10}$  CFU/mL) of *B/l* 1821L and *B/l* 1951 with/without treatment of crude *B/l* 1821L putative antibacterial proteins (ABPs) after incubation at 30°C over 24 hours.



**Figure S8.** Effect of crude *B/l* 1821 putative antibacterial proteins (ABPs) on the OD<sub>600nm</sub> reading of *B/l* 1821L and *B/l* 1951 after incubation at 30°C for various time intervals.

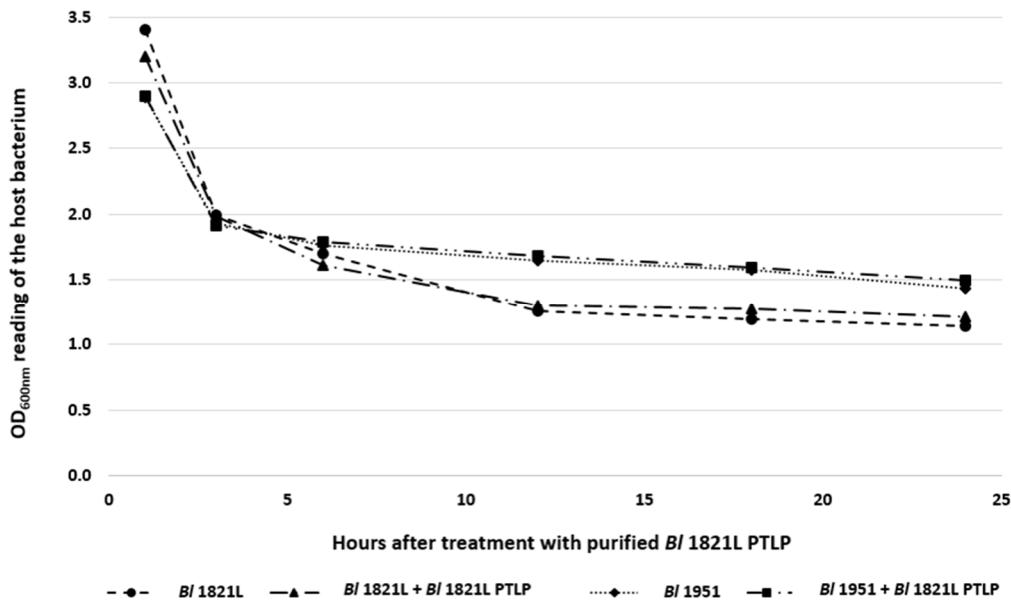
**Table S5. Effect of purified *B*/1821L putative ~48 kD phage tail-like protein (PTLP) on the number of viable cells of *B*/1821L and *B*/1951 after incubation at 30°C for various time intervals. Data presents the mean values of one experiment. Values of % decrease/increase in the number of viable cells are calculated from CFUs values of corresponding time intervals .**

| Time intervals<br>(Hours) | <i>B</i> /1821L      | <i>B</i> /1821L<br>+<br><i>B</i> /1821L PTLP | % Decrease/increase<br>in no. of viable cells | <i>B</i> /1951      | <i>B</i> /1951<br>+<br><i>B</i> /1821L PTLP | % Decrease/increase<br>in no. of viable cells |
|---------------------------|----------------------|--|---|---------------------|---|---|
| 1                         | 1.74E+07<br>(7.241)* | 2.81E+07<br>(7.448)                          | -61.21  | 9.05E+06<br>(6.957) | 1.66E+07<br>(7.220)                         | -83.43  |
| 3                         | 2.41E+07<br>(7.381)  | 2.16E+07<br>(7.334)                          | 10.19   | 6.20E+06<br>(6.792) | 8.80E+06<br>(6.944)                         | -41.94  |
| 6                         | 1.13E+07<br>(7.051)  | 1.64E+07<br>(7.215)                          | -45.78  | 1.17E+07<br>(7.066) | 1.08E+07<br>(7.031)                         | 7.73  |
| 12                        | 2.34E+07<br>(7.369)  | 2.14E+07<br>(7.330)                          | 8.55  | 2.58E+07<br>(7.411) | 2.01E+07<br>(7.302)                         | 22.14   |
| 18                        | 1.68E+07<br>(7.224)  | 2.78E+07<br>(7.443)                          | -65.67  | 2.37E+07<br>(7.375) | 1.56E+07<br>(7.193)                         | 34.18   |
| 24                        | 2.33E+07<br>(7.366)  | 2.87E+07<br>(7.457)                          | -23.23  | 1.87E+07<br>(7.271) | 2.03E+07<br>(7.307)                         | -8.85   |

\*= The values in parenthesis indicate the number of viable cells (CFUs/mL) converted into log<sub>10</sub> CFU/mL.

**Table S6.** Effect of purified *B/ 1821L* ~48 kD putative phage tail-like protein (PTLP) on the OD<sub>600nm</sub> reading of *B/ 1821L* and *B/ 1951* after incubation at 30°C for various time intervals. Data presents the mean values of one experiment.

| Time intervals<br>(Hours)   | <i>B/ 1821L</i> | <i>B/ 1821L</i><br>+<br><i>B/ 1821L</i> PTLP | % Decrease/increase<br>in OD <sub>600nm</sub> reading | <i>B/ 1951</i> | <i>B/ 1951</i><br>+<br><i>B/ 1821L</i> PTLP | % Decrease/increase<br>in OD <sub>600nm</sub> reading |
|---|-----------------|--|---|----------------|---|---|
| 1   | 3.40            | 3.20   | 5.88  | 2.89           | 2.90  | -0.35   |
| 3   | 1.99            | 1.98   | 0.50  | 1.93           | 1.91  | 1.04  |
| 6   | 1.70            | 1.61   | 5.01  | 1.76           | 1.79  | -1.70   |
| 12  | 1.26            | 1.30   | -3.59   | 1.65           | 1.68  | -1.82   |
| 18  | 1.19            | 1.27   | -6.30   | 1.58           | 1.59  | -0.63   |
| 24  | 1.14            | 1.21   | -6.14   | 1.43           | 1.50  | -4.90   |
| <b>% Decrease from<br/>the start (1 hour) to<br/>the end (24 hours)<br/>of incubation</b> | <b>66.5%</b>    | <b>62.2%</b>                                 |   |                | <b>50.5%</b>                                | <b>48.3%</b>  |



**Figure S9.** Effect of purified *B/ 1821* putative phage tail-like protein (~48 kD) on the OD<sub>600nm</sub> reading of *B/ 1821L* and *B/ 1951* after incubation at 30°C for various time intervals.

**Table S7.** Phage-like element PBSX protein XkdK and phage tail-sheath proteins from different gram-positive bacteria.

| Accession   | Protein name                 | Organism  |
|-------------|------------------------------|---|
| A0A0D1WNL8  | Phage tail sheath            | <i>Aneurinibacillus migulanus</i>                             |
| A0A410KN98I | Phage tail sheath            | <i>Bacillus aerophilus</i>                                    |
| R4JQA6      | Structural protein           | <i>Bacillus</i> phage PBP180                                  |
| A0A5B0B6Z4  | Phage-like element PBSX XkdK | <i>Bacillus</i> sp. ANT_WA51                                  |
| A0A410QZ71  | Phage-like element PBSX XkdK | <i>Bacillus</i> sp. WR11                                      |
| A0A6H0H1P2  | Phage-like element PBSX XkdK | <i>Bacillus subtilis</i> subsp. <i>subtilis</i> str. SMY      |
| P54331      | Phage-like element PBSX XkdK | <i>Bacillus subtilis</i> 168                                  |
| C0Z5G9      | Uncharacterised protein      | <i>Brevibacillus brevis</i> (strain 47/JCM 6285/ NBRC 100599) |
| A0A3M8B733  | Phage tail                   | <i>Brevibacillus gelatinii</i>                                |
| A0A075R9L5  | Phage tail sheath            | <i>Brevibacillus laterosporus</i> LMG 15441                   |
| Q18BN0      | Phage-like element PBSX XkdK | <i>Clostridioides difficile</i> 630                           |
| A0A061P351  | Phage-like element PBSX XkdK | <i>Geomicrobium</i> sp. JCM 19039                             |

**Figure S10. Amino acids alignment of identified *B/ 1821L* and *B/ 1951* phage like-element PBSX protein XkdK (A0A518VEB0) with the similar proteins of other gram-positive bacteria (See SM Table 7) using the programme CLUSTALO.**

|            |                  |     |  |     |
|------------|------------------|-----|--|-----|
| AOA0D1WNL8 | AOA0D1WNL8_ANEMI | 1   | -----MAGGTQGERKVRPGFYARFISAAQDRIAVAPRGTVILPLTLNWGRAKE          | 50  |
| AOA3M8B733 | AOA3M8B733_9BACL | 1   | -----MTIQRERPGVTVELIAKAKERV-VPKSGVVLVPYQAEWGAPDE               | 42  |
| AOA5B0B624 | AOA5B0B624_9BACI | 1   | -----MNGGTFTTGKEKERAGIYFNFKTTAQERVSLSERGTVALPVASSWGEAKT        | 50  |
| AOA6HOH1P2 | AOA6HOH1P2_BACIU | 1   | -----MNGGTFTTGKEKERAGIYFNFKTTAQERVSLSERGTVALPVASSWGEAKT        | 50  |
| AOA061P351 | AOA061P351_9BACL | 1   | -----MNGGTFAFPGVKEKERAGIYFRFTAANDRLSVPGERGTVALPLESWGAPKT       | 50  |
| AOA075R9L5 | AOA075R9L5_BRELA | 1   | MKNKYDNTYLHFEEVNSMTIQRERPGVNVELAKAKAQRERV-LPKSGVVLVPYLAEGWAPDQ | 59  |
| AOA410KN98 | AOA410KN98_9BACI | 1   | -----MNGGTFTPGTEKKRPGIYFNFKTTAEQRITLGERGTVALPLVMSWGEPKT        | 50  |
| AOA410QZ71 | AOA410QZ71_9BACI | 1   | -----MNGGTFTTGKEKERAGIYFNFKTTAQERVSLSERGTVALPVASSWGEAKT        | 50  |
| AOA518VEB0 | AOA518VEB0_BRELA | 1   | -----MTIQRERPGVNVELAKAKAQRERV-LPKSGVVLVPYLAEGWAPDQ             | 42  |
| C0Z5G9     | C0Z5G9_BREBN     | 1   | -----MTIQRERPGVTVELIAKAKERV-VPKSGVVLVPYQAEWGAPDE               | 42  |
| Q18BNO     | Q18BNO_CLOD6     | 1   | -----  | 0   |
| R4JQA6     | R4JQA6_9CAUD     | 1   | -----MNGGTFTPGTEKKRPGIYFNFKTTAEQRITLGERGTVALPLVMSWGEPKT        | 50  |
| P54331     | XKDK_BACSU       | 1   | -----MNGGTFTTGKEKERAGIYFNFKTTAQERVSLSERGTVALPVASSWGEAKT        | 50  |
| AOA0D1WNL8 | AOA0D1WNL8_ANEMI | 51  | FTTIEVEKDMDKLGYDYNDPEMLLIREARKLAKKVVKVYKLNKGAKAGTFGTT-----     | 104 |
| AOA3M8B733 | AOA3M8B733_9BACL | 43  | LVKLGSYEERMTEF-----GKVDTVELAAEGGATILAYRMTNGNASKAVYEQA-----     | 91  |
| AOA5B0B624 | AOA5B0B624_9BACI | 51  | FVSISSVEDLNKKVGLSIDDPSSLLLREAKKNAKTVLMLYRILTEGVVRASADIA-----   | 102 |
| AOA6HOH1P2 | AOA6HOH1P2_BACIU | 51  | FVSISSVEDLNKKVGLSIDDPSSLLLREAKKNAKTVLMLYRILTEGVVRASADIA-----   | 102 |
| AOA061P351 | AOA061P351_9BACL | 51  | FVEINGPDDVLKKYGLSVNDSSVLLKEAMKRSQTIVLAYRNEGSKAETVIGESSGGGS     | 110 |
| AOA075R9L5 | AOA075R9L5_BRELA | 60  | VITMKGYEERVAETF-----GQIDILELAAEGGATVVGYRMTNGKSVAASYSQE-----    | 108 |
| AOA410KN98 | AOA410KN98_9BACI | 51  | FISVSDMEDLNKKVGLNIDDKSLLLREAKKKAQTVLVLYRILNEGEPAKAEIA-----     | 102 |
| AOA410QZ71 | AOA410QZ71_9BACI | 51  | FVSISSVEDLNKKVGLSIDDPSSLLLREAKKNAKTVLMLYRILTEGVVRASADIA-----   | 102 |
| AOA518VEB0 | AOA518VEB0_BRELA | 43  | VITMKGYEERVAETF-----GHGILELAAEGGATVLGYRMTSGNGVAASYTQS-----     | 91  |
| C0Z5G9     | C0Z5G9_BREBN     | 43  | LVKLGSFEERIAQTF-----GKVDTVELAAEGGATILAYRMTNGTATKAAYEQA-----    | 91  |
| Q18BNO     | Q18BNO_CLOD6     | 1   | -----MAIGLPSINI-----   | 10  |
| R4JQA6     | R4JQA6_9CAUD     | 51  | FISVSDMEDLNKKVGLNIDDKSLLLREAKKKAQTVLVLYRILNEGEPAKAEIA-----     | 102 |
| P54331     | XKDK_BACSU       | 51  | FVSISSVEDLNKKVGLSIDDPSSLLLREAKKNAKTVLMLYRILTEGVVRASADIA-----   | 102 |
| AOA0D1WNL8 | AOA0D1WNL8_ANEMI | 105 | -----SICTVEIINDGTRGNDITIVSQNVLDTTKDVITYVKGRQVDKQTQ             | 151 |
| AOA3M8B733 | AOA3M8B733_9BACL | 92  | -----DAIRIELYPGLLGNDLRIVISASTSEPGKKELQVKG-PLQTEKFNF            | 137 |
| AOA5B0B624 | AOA5B0B624_9BACI | 103 | -----EGVKATEVYGGTKGNDIIIRINQNVLNDANSFDVTIYMDSEVDKQTV           | 149 |
| AOA6HOH1P2 | AOA6HOH1P2_BACIU | 103 | -----EGVKATEVYGGTKGNDIIIRINQNVLNDANSFDVTIYMDSEVDKQTV           | 149 |
| AOA061P351 | AOA061P351_9BACL | 111 | DGDDEEEGASGQSNGLRAVEKFGGMKGNDIQIRVSENVLDSLELFDTIYLNNVAVNRQSV   | 170 |
| AOA075R9L5 | AOA075R9L5_BRELA | 109 | -----GSIAIQARYPGLVGNELQISIKDSTAELGKKELQVKG-PIKTEKFNF           | 154 |
| AOA410KN98 | AOA410KN98_9BACI | 103 | -----ENFVVTINYGGQKGNEITIQLVAENVLDSTKRDVITYLGTDIVDKQVV          | 149 |
| AOA410QZ71 | AOA410QZ71_9BACI | 103 | -----EGVKATEVYGGTKGNDIIIRINQNVLNDANSFDVTIYMDSEVDRQTV           | 149 |
| AOA518VEB0 | AOA518VEB0_BRELA | 92  | -----DSFTLERYPGLVGNDLQISIKESTVELGKKELQVKG-PIKTEKFNF            | 137 |
| C0Z5G9     | C0Z5G9_BREBN     | 92  | -----DAIRIELYPGLVGNELKVTITVSTSEPGKKELQVTG-PLQTEKFNF            | 137 |
| Q18BNO     | Q18BNO_CLOD6     | 11  | -----SFKELE-----TTVKERSARGIIAMVLKDAKALGLNEIHEKEDI              | 49  |
| R4JQA6     | R4JQA6_9CAUD     | 103 | -----ENFVVTINYGGQKGNEITIQLVAENVLDSTKRDVITYLGTDIVDKQVV          | 149 |
| P54331     | XKDK_BACSU       | 103 | -----EGVKATEVYGGTKGNDIIIRINQNVLNDANSFDVTIYMDSEVDKQTV           | 149 |



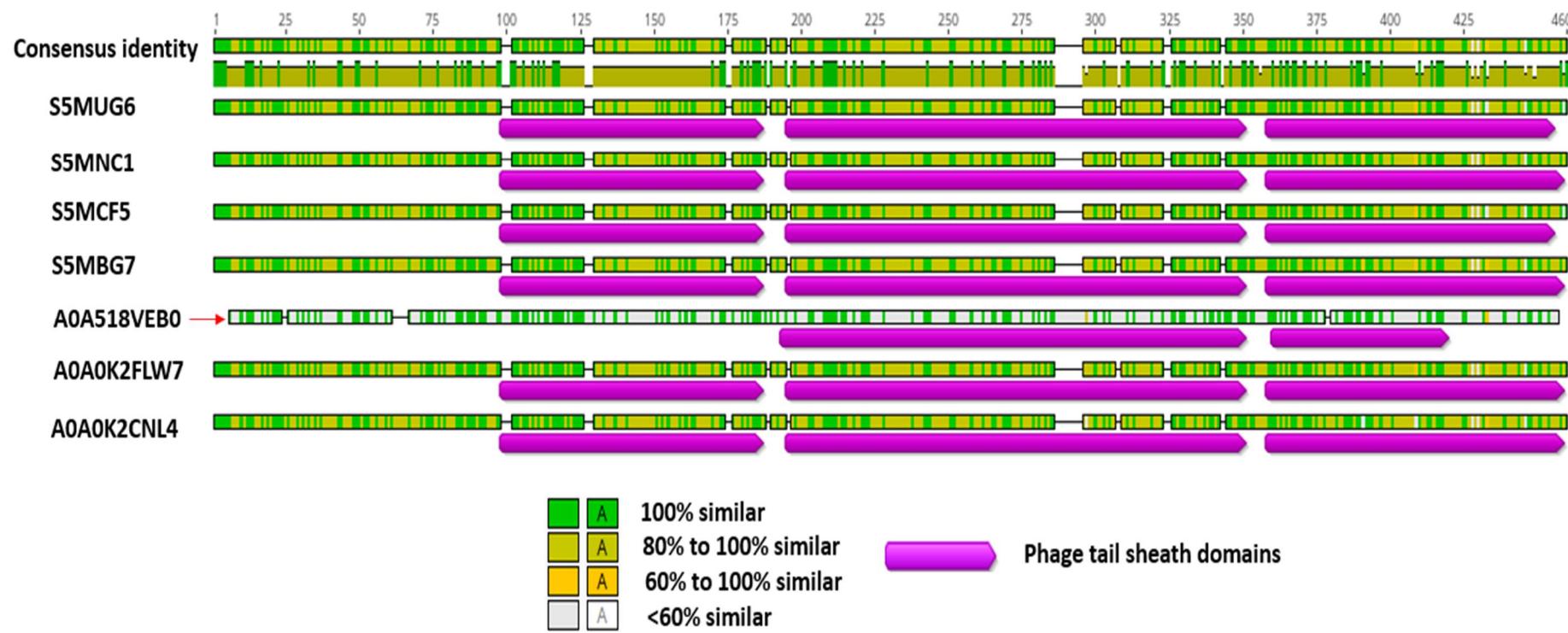
\*Highlighted dark grey color amino acids denote similarity among the aligned phage-like element PBSX protein XkdK.  
 (Refer to SM Tables S8 & S9)

**Table S8.** Distance matrices of identified *B. 1821L* and *B. 1951* phage-like element PBSX protein XkdK (A0A518VEB0) with the similar proteins of other gram-positive bacteria.

| Uniprot Accession #<br>(www.uniprot.org) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |            |            |            |            |            |            |            |            |        |        |        |        |
|--|--|------------|------------|------------|------------|------------|------------|------------|------------|--------|--------|--------|--------|
|  | A0A0D1WNL8   | A0A3M8B733 | A0A5B0B6Z4 | A0A6H0H1P2 | A0A061P351 | A0A075R9L5 | A0A410KN98 | A0A410QZ71 | A0A518VEB0 | C0Z5G9 | Q18BN0 | R4JQA6 | P54331 |
| <b>A0A0D1WNL8</b>                        | 1.34   | 0.8        | 0.8        | 0.84       | 1.4        | 0.8        | 0.8        | 0.8        | 1.33       | 1.46   | 1.51   | 0.8    | 0.8    |
| <b>A0A3M8B733</b>                        | 1.34   |            | 1.44       | 1.44       | 1.43       | 0.38       | 1.4        | 1.44       | 0.36       | 0.14   | 1.64   | 1.4    | 1.44   |
| <b>A0A5B0B6Z4</b>                        | 0.8  | 1.44       |            | 0          | 0.56       | 1.52       | 0.37       | 0          | 1.45       | 1.45   | 1.49   | 0.37   | 0      |
| <b>A0A6H0H1P2</b>                        | 0.8  | 1.44       | 0          |            | 0.56       | 1.52       | 0.37       | 0          | 1.45       | 1.45   | 1.49   | 0.37   | 0      |
| <b>A0A061P351</b>                        | 0.84   | 1.43       | 0.56       | 0.56       |            | 1.42       | 0.57       | 0.56       | 1.45       | 1.38   | 1.54   | 0.57   | 0.56   |
| <b>A0A075R9L5</b>                        | 1.4  | 0.38       | 1.52       | 1.52       | 1.42       |            | 1.44       | 1.55       | 0.1        | 0.39   | 1.49   | 1.44   | 1.55   |
| <b>A0A410KN98</b>                        | 0.8  | 1.4        | 0.37       | 0.37       | 0.57       | 1.44       |            | 0.37       | 1.4        | 1.37   | 1.66   | 0      | 0.37   |
| <b>A0A410QZ71</b>                        | 0.8  | 1.44       | 0          | 0          | 0.56       | 1.55       | 0.37       |            | 1.45       | 1.45   | 1.49   | 0.37   | 0      |
| <b>A0A518VEB0</b>                        | 1.33   | 0.36       | 1.45       | 1.45       | 1.45       | 0.1        | 1.4        | 1.45       |            | 0.39   | 1.52   | 1.4    | 1.45   |
| <b>C0Z5G9</b>                            | 1.46   | 0.14       | 1.45       | 1.45       | 1.38       | 0.39       | 1.37       | 1.45       | 0.39       |        | 1.66   | 1.37   | 1.45   |
| <b>Q18BN0</b>                            | 1.51   | 1.64       | 1.49       | 1.49       | 1.54       | 1.49       | 1.66       | 1.49       | 1.52       | 1.66   |        | 1.66   | 1.49   |
| <b>R4JQA6</b>                            | 0.8  | 1.4        | 0.37       | 0.37       | 0.57       | 1.44       | 0          | 0.37       | 1.4        | 1.37   | 1.66   |        | 0.37   |
| <b>P54331</b>                            | 0.8  | 1.44       | 0          | 0          | 0.56       | 1.55       | 0.37       | 0          | 1.45       | 1.45   | 1.49   |        | 0.37   |

**Table S9.** Amino acids alignment % of identified *B*/1821L and *B*/1951 phage-like element PBSX protein Xkdk (A0A518VEB0) with the similar proteins of other gram-positive bacteria.

| Uniprot Accession #<br>( <a href="https://www.uniprot.org">www.uniprot.org</a> ) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |            |            |            |            |            |            |            |            |        |        |        |
|--|--|------------|------------|------------|------------|------------|------------|------------|------------|--------|--------|--------|
|  | A0A0D1WNL8   | A0A3M8B733 | A0A5B0B6Z4 | A0A6H0H1P2 | A0A061P351 | A0A075R9L5 | A0A410KN98 | A0A410QZ71 | A0A518VEB0 | C0Z5G9 | Q18BN0 | R4JQA6 |
| <b>A0A0D1WNL8</b>  | 23.9   | 41.5       | 41.5       | 39.8       | 22.0       | 42.6       | 41.5       | 22.7       | 23.2       | 22.0   | 42.6   | 41.5   |
| <b>A0A3M8B733</b>  | 23.9   | 21.2       | 21.2       | 20.7       | 68.2       | 20.6       | 21.2       | 69.0       | 86.0       | 18.4   | 20.6   | 21.2   |
| <b>A0A5B0B6Z4</b>  | 41.5   | 21.2       | 100.0      | 51.8       | 20.3       | 64.8       | 100.0      | 21.2       | 20.5       | 22.4   | 64.8   | 100.0  |
| <b>A0A6H0H1P2</b>  | 41.5   | 21.2       | 100.0      | 51.8       | 20.3       | 64.8       | 100.0      | 21.2       | 20.5       | 22.4   | 64.8   | 100.0  |
| <b>A0A061P351</b>  | 39.8   | 20.7       | 51.8       | 51.8       | 19.0       | 53.4       | 51.8       | 18.8       | 19.4       | 20.5   | 53.4   | 51.8   |
| <b>A0A075R9L5</b>  | 22.0   | 68.2       | 20.3       | 20.3       | 19.0       | 19.8       | 20.3       | 89.1       | 67.4       | 19.3   | 19.8   | 20.3   |
| <b>A0A410KN98</b>  | 42.6   | 20.6       | 64.8       | 64.8       | 53.4       | 19.8       | 64.8       | 20.3       | 22.0       | 20.1   | 100.0  | 64.8   |
| <b>A0A410QZ71</b>  | 41.5   | 21.2       | 100.0      | 100.0      | 51.8       | 20.3       | 64.8       | 21.2       | 20.5       | 22.4   | 64.8   | 100.0  |
| <b>A0A518VEB0</b>  | 22.7   | 69.0       | 21.2       | 21.2       | 18.8       | 89.1       | 20.3       | 21.2       | 68.1       | 19.9   | 20.3   | 21.2   |
| <b>C0Z5G9</b>  | 23.2   | 86.0       | 20.5       | 20.5       | 19.4       | 67.4       | 22.0       | 20.5       | 68.1       | 20.2   | 22.0   | 20.5   |
| <b>Q18BN0</b>  | 22.0   | 18.4       | 22.4       | 22.4       | 20.5       | 19.3       | 20.1       | 22.4       | 19.9       | 20.2   | 20.1   | 22.4   |
| <b>R4JQA6</b>  | 42.6   | 20.6       | 64.8       | 64.8       | 53.4       | 19.8       | 100.0      | 64.8       | 20.3       | 22.0   | 20.1   | 64.8   |
| <b>P54331</b>  | 41.5   | 21.2       | 100.0      | 100.0      | 51.8       | 20.3       | 64.8       | 100.0      | 21.2       | 20.5   | 22.4   | 64.8   |



**Figure S11.**Amino acids alignment and percentage identity of identified putative phage tail-sheath protein (AOA518VEB0, shown with red arrow) of *BI* 1821L and *BI* 1951 with similar proteins of different *BI* phages including Abouo (S5MUG6), Jimmer1 (S5MNC1), Davies (S5MCF5), Jimmer2, (S5MBG7), Powder (AOAOK2FLW7), and Osiris (AOAOK2CNL4) using Geneious basic.

(Refer to SM Figure S12 & Tables S10 and S11)

**Figure S12. Amino acids alignment of identified *B*/1821L and *B*/1951 putative phage tail-sheath protein (A0A518VEB0) with the phage tail-sheath protein of different *B*/ phages using the programme CLUSTALO.**

|            |                  |     |   |     |
|------------|------------------|-----|---|-----|
| A0A518VEB0 | A0A518VEB0_BRELA | 1   | -----MTIQRERPGVNVELKAKA-QERVLPKSGVVLPYLAEGAPDQVITMKGYEERVA    | 54  |
| S5MUG6     | S5MUG6_9CAUD     | 1   | MMQNWIQAQNVRPGTYVNVSSEAKPQGAISERGIVTMALSLSWGPSGEVIEIQAGENTLD  | 60  |
| S5MNC1     | S5MNC1_9CAUD     | 1   | MMQNWIQAQNVRPGTYVNVSSEAKPQGAISERGIVTMALSLSWGPSGEVIEIQAGENTLD  | 60  |
| S5MCF5     | S5MCF5_9CAUD     | 1   | MMQNWIQAQNVRPGTYVNVSSEAKPQGAISERGIVTMALSLSWGPSGEVIEIQAGENTLD  | 60  |
| S5MBG7     | S5MBG7_9CAUD     | 1   | MMQNWIQAQNVRPGTYVNVSSEAKPQGAISERGIVTMALSLSWGPSGEVIEIQAGENTLD  | 60  |
| A0A0K2FLW7 | A0A0K2FLW7_9CAUD | 1   | MMQNWIQAQNVRPGTYVNVSSEAKPQGAISERGIVTMALSLSWGPSGEVIEIQAGENTLD  | 60  |
| A0A0K2CNL4 | A0A0K2CNL4_9CAUD | 1   | MMQNWIQAQNVRPGTYVNVSSEAKPQGAISERGIVTMALSLSWGPSGEVIEIQAGENTLD  | 60  |
|            |                  |     | : : : *.*. *:: :* : . : : *.* : .** .:*** :.. * : :           |     |
| A0A518VEB0 | A0A518VEB0_BRELA | 55  | ETF-----GHIGILELPAEGGATVIGYRMTSGNGVAASYTQSDFSTLEARYPGIVGNDLQ  | 109 |
| S5MUG6     | S5MUG6_9CAUD     | 61  | KLGYDISEPQILLVQEALKRAKTLLYRINTGSKAQ---ASSDNLTVTALYGGVRGNDIT   | 117 |
| S5MNC1     | S5MNC1_9CAUD     | 61  | KLGYDISEPQILLVQEALKRAKTLLYRINTGSKAQ---ASSDNLTVTALYGGVRGNDIT   | 117 |
| S5MCF5     | S5MCF5_9CAUD     | 61  | KLGYDISEPQILLVQEALKRAKTLLYRINTGSKAQ---ASSDNLTVTALYGGVRGNDIT   | 117 |
| S5MBG7     | S5MBG7_9CAUD     | 61  | KLGYDISEPQILLVQEALKRAKTLLYRINTGSKAQ---ASSDNLTVTALYGGVRGNDIT   | 117 |
| A0A0K2FLW7 | A0A0K2FLW7_9CAUD | 61  | KLGYDISEPQILLVQEALKRAKTLLYRINTGSKAQ---ASSDNLTVTALYGGVRGNDIT   | 117 |
| A0A0K2CNL4 | A0A0K2CNL4_9CAUD | 61  | KLGYDISEPQILLVQEALKRAKTLLYRINTGSKAQ---ASSDNLTVTALYGGVRGNDIT   | 117 |
|            |                  |     | : :* : :* : . *;* **: :.. . : .**.:*: * * * : ***:            |     |
| A0A518VEB0 | A0A518VEB0_BRELA | 110 | ISIKESTVELGKK--ELQVKGP KTEKF SFANVDELVTKAEQSIYIKVKKLGDKVAEETM | 167 |
| S5MUG6     | S5MUG6_9CAUD     | 118 | VVVEQNIDDETTFIVKTLVAGSIVDNQLA-----KKIEDLKANKFVTSGTGALV--ASA   | 170 |
| S5MNC1     | S5MNC1_9CAUD     | 118 | VVVEQNIDDETTFIVKTLVAGSIVDNQLA-----KKIEDLKANKFVTSGTGALV--ASA   | 170 |
| S5MCF5     | S5MCF5_9CAUD     | 118 | VVVEQNIDDETTFIVKTLVAGSIVDNQLA-----KKIEDLKANKFVTSGTGALV--ASA   | 170 |
| S5MBG7     | S5MBG7_9CAUD     | 118 | VVVEQNIDDETTFIVKTLVAGSIVDNQLA-----KKIEDLKANKFVTSGTGALV--ASA   | 170 |
| A0A0K2FLW7 | A0A0K2FLW7_9CAUD | 118 | VVVEQNIDDETTFIVKTLVAGSIVDNQLA-----KKIEDLKANKFVTSGTGALV--ASA   | 170 |
| A0A0K2CNL4 | A0A0K2CNL4_9CAUD | 118 | VVVEQNIDDETTFIVKTLVAGSIVDNQLA-----KKIEDLKANKFVTSGTGALV--ASA   | 170 |
|            |                  |     | : : : . : . : * * * : : : : : : : : * * : :                   |     |
| A0A518VEB0 | A0A518VEB0_BRELA | 168 | MISIEGGITSGITSLTANDFTTLFNSISGTDFDAMYLPADVAIQAAAKQEMSDRELFSKK  | 227 |
| S5MUG6     | S5MUG6_9CAUD     | 171 | GIPITGGIDGTEIG--VNHTAYREAIELHDFDAMAVFYDDPTIKSVYVAFAKRLANQQNR  | 228 |
| S5MNC1     | S5MNC1_9CAUD     | 171 | GIPITGGIDGTEIG--VNHTAYREAIELHDFDAMAVFYDDPTIKSVYVAFAKRLANQQNR  | 228 |
| S5MCF5     | S5MCF5_9CAUD     | 171 | GIPITGGIDGTEIG--VNHTAYREAIELHDFDAMAVFYDDPTIKSVYVAFAKRLANQQNR  | 228 |
| S5MBG7     | S5MBG7_9CAUD     | 171 | GIPITGGIDGTEIG--VNHTAYREAIELHDFDAMAVFYDDPTIKSVYVAFAKRLANQQNR  | 228 |
| A0A0K2FLW7 | A0A0K2FLW7_9CAUD | 171 | GIPITGGIDGTEIG--VNHTAYREAIELHDFDAMAVFYDDPTIKSVYVAFAKRLANQQNR  | 228 |
| A0A0K2CNL4 | A0A0K2CNL4_9CAUD | 171 | GIPITGGIDGTEIG--VNHTAYREAIELHDFDAMAVFYDDPTIKSVYVAFAKRLANQQNR  | 228 |
|            |                  |     | * * *.*. * : : * : . * * * : * : * : : . * . : :              |     |

|            |                  |       |     |   |     |
|------------|------------------|-------|-----|---|-----|
| AOA518VEB0 | AOA518VEB0       | BRELA | 228 | RSTLVIGGLKDNNMNEHVQRSVANNNSRRVNCAIAGQHVGKTYGSLEWAALAGMIA                        | 287 |
| S5MUG6     | S5MUG6_9CAUD     |       | 229 | FIQIVVPNYAQADDPT-----VISVSGVILSNSTVIDAVKATAWVAGATA                              | 274 |
| S5MNC1     | S5MNC1_9CAUD     |       | 229 | FIQIVVPNYAQADDPT-----VISVSGVILSNSTVIDAVKATAWVAGATA                              | 274 |
| S5MCF5     | S5MCF5_9CAUD     |       | 229 | FIQIVVPNYAQADDPT-----VISVSGVILSNSTVIDAVKATAWVAGATA                              | 274 |
| S5MBG7     | S5MBG7_9CAUD     |       | 229 | FIQIVVPNYAQADDPT-----VISVSGVILSNSTVIDAVKATAWVAGATA                              | 274 |
| AOAOK2FLW7 | AOAOK2FLW7_9CAUD |       | 229 | FIQIVVPNYAQADDPT-----VISVSGVILSNSTVIDAVKATAWVAGATA                              | 274 |
| AOAOK2CNL4 | AOAOK2CNL4_9CAUD |       | 229 | FIQIVVPNYAQADDPT-----VISVSGVILSNSTVIDAVKATAWVAGATA                              | 274 |
|            |                  |       |     | : * : . : * : * : . : * : . : * : . : * : * : * : * : * :                       |     |
| AOA518VEB0 | AOA518VEB0       | BRELA | 288 | ATPAHISLSAQLVPL-KKAEDWGHTDQAAINSGTLIAVRDGDVYLIIESAVNLTTLKA                      | 346 |
| S5MUG6     | S5MUG6_9CAUD     |       | 275 | GANVNQLTHTAYDDAVAVHGRNLNDSQITKALLNGEFLFELHNGKVVEQDINTFTSFSP                     | 334 |
| S5MNC1     | S5MNC1_9CAUD     |       | 275 | GANVNQLTHTAYDDAVAVHGRNLNDSQITKALLNGEFLFELHNGKVVEQDINTFTSFSP                     | 334 |
| S5MCF5     | S5MCF5_9CAUD     |       | 275 | GANVNQLTHTAYDDAVAVHGRNLNDSQITKALLNGEFLFELHNGKVVEQDINTFTSFSP                     | 334 |
| S5MBG7     | S5MBG7_9CAUD     |       | 275 | GANVNQLTHTAYDDAVAVHGRNLNDSQITKALLNGEFLFELHNGKVVEQDINTFTSFSP                     | 334 |
| AOAOK2FLW7 | AOAOK2FLW7_9CAUD |       | 275 | GANVNQLTHTAYDDAVAVHGRNLNDSQITKALLNGEFLFELHNGKVVEQDINTFTSFSP                     | 334 |
| AOAOK2CNL4 | AOAOK2CNL4_9CAUD |       | 275 | GANANQSLTHTAYDDAVAVHGRNLNDSQITKALLNGEFLFELHNGKVVEQDINTFTSFNP                    | 334 |
|            |                  |       |     | : . : * : . : . : * : * : * : . : . : * : . : * : . : * : * : * : . : * : * : : |     |
| AOA518VEB0 | AOA518VEB0       | BRELA | 347 | AEREDFGKIRVSMILDQIVNDIT-SVGKKYKGKLDNNDIGGATFVGAVKTYLEVREAQGA                    | 405 |
| S5MUG6     | S5MUG6_9CAUD     |       | 335 | DKRKHFSKNRVVRITINGITKDWKLAFDEQYLGKGDNADGRNLYKKECIKISEQYQAMGA                    | 394 |
| S5MNC1     | S5MNC1_9CAUD     |       | 335 | DKRKHFSKNRVVRITINGITKDWKLAFDEQYLGKGDNADGRNLYKKECIKISEQYQAMGA                    | 394 |
| S5MCF5     | S5MCF5_9CAUD     |       | 335 | DKRKHFSKNRVVRITINGITKDWKLAFDEQYLGKGDNADGRNLYKKECIKISEQYQAMGA                    | 394 |
| S5MBG7     | S5MBG7_9CAUD     |       | 335 | DKRKHFSKNRVVRITINGITKDWKLAFDEQYLGKGDNADGRNLYKKECIKISEQYQAMGA                    | 394 |
| AOAOK2FLW7 | AOAOK2FLW7_9CAUD |       | 335 | DKRKHFSKNRVVRITINGITKDWKLAFDEQYLGKGDNADGRNLYKKECIKISEQYQAMGA                    | 394 |
| AOAOK2CNL4 | AOAOK2CNL4_9CAUD |       | 335 | DKRKHFSKNRVVRITINGITKDWKLAFDEQYLGKYDNADGRNLYKKECIKIVEYQAMGA                     | 394 |
|            |                  |       |     | : * : . * : * : * : . : * : . : * : * : * : * : . : * : . : * : * : * : :       |     |
| AOA518VEB0 | AOA518VEB0       | BRELA | 406 | IDQGWIFEDKKN---GIGDKRGFRIAAKPLDAIEIFDIEWEVL---                                  | 445 |
| S5MUG6     | S5MUG6_9CAUD     |       | 395 | IQN---FDAQKDIIVLPGEDSDSLLTEGYIQAVDSMEKNYLKAVAR                                  | 437 |
| S5MNC1     | S5MNC1_9CAUD     |       | 395 | IQN---FDAQKDIIVSPGNDSDSLITEGYIQPVDAMEKNYLKAVVR                                  | 437 |
| S5MCF5     | S5MCF5_9CAUD     |       | 395 | IQN---FDAQKDIIVLPGEDSDSLLTEGYIQAVDSMEKNYLKAVAR                                  | 437 |
| S5MBG7     | S5MBG7_9CAUD     |       | 395 | IQN---FDAQKDIIVSPGNDSDSLITEGYIQPVDAMEKNYLKAVVR                                  | 437 |
| AOAOK2FLW7 | AOAOK2FLW7_9CAUD |       | 395 | IQN---FDAQKDIIVSPGNDSDSLITEGYIQPVDAMEKNYLKAVVR                                  | 437 |
| AOAOK2CNL4 | AOAOK2CNL4_9CAUD |       | 395 | IQN---FDAQKDIIVSPGNDSDSLITEGYIQPVDAMEKNYLKAVVR                                  | 437 |
|            |                  |       |     | * : * : * : * : * : . : * : * : . : * : * : . : * : * : * : :                   |     |

\*Highlighted dark grey color amino acids denote similarity among the aligned phage tail-sheath like proteins.  
 (Refer to SM Tables S10 & S11)

**Table S10.** Distance matrices of identified *B/ 1821L* and *B/ 1951* putative phage tail-sheath protein (A0A518VEB0) with the phage tail-sheath proteins of different *B/* phages .

| Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |        |        |        |            |            |            |
|--|--|--------|--------|--------|------------|------------|------------|
|  | S5MUG6   | S5MNC1 | S5MCF5 | S5MBG7 | A0A518VEB0 | A0A0K2FLW7 | A0A0K2CNL4 |
| <b>S5MUG6</b>  |  | 0.01   | 0      | 0.01   | 1.56       | 0.01       | 0.03       |
| <b>S5MNC1</b>  | 0.01   |        | 0.01   | 0      | 1.51       | 0          | 0.01       |
| <b>S5MCF5</b>  | 0  | 0.01   |        | 0.01   | 1.56       | 0.01       | 0.03       |
| <b>S5MBG7</b>  | 0.01   | 0      | 0.01   |        | 1.51       | 0          | 0.01       |
| <b>A0A518VEB0</b>  | 1.56   | 1.51   | 1.56   | 1.51   |            | 1.51       | 1.52       |
| <b>A0A0K2FLW7</b>  | 0.01   | 0      | 0.01   | 0      | 1.51       |            | 0.01       |
| <b>A0A0K2CNL4</b>  | 0.03   | 0.01   | 0.03   | 0.01   | 1.52       | 0.01       |            |

**Table S11.** Amino acids alignment (%) of identified *B*/*I* 1821L and *B*/*I* 1951 putative phage tail-sheath protein (A0A518VEB0) with the phage tail-sheath proteins of different *B*/*I* phages.

| Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |        |        |        |            |            |            |
|--|--|--------|--------|--------|------------|------------|------------|
|  | S5MUG6   | S5MNC1 | S5MCF5 | S5MBG7 | A0A518VEB0 | A0A0K2FLW7 | A0A0K2CNL4 |
| <b>S5MUG6</b>  | 98.6   | 100    | 98.6   | 21.9   | 98.6       | 97.5       |            |
| <b>S5MNC1</b>  | 98.6   |        | 98.6   | 100    | 21.7       | 100        | 98.9       |
| <b>S5MCF5</b>  | 100  | 98.6   |        | 98.6   | 21.9       | 98.6       | 97.5       |
| <b>S5MBG7</b>  | 98.6   | 100    | 98.6   |        | 21.7       | 100        | 98.9       |
| <b>A0A518VEB0</b>  | 21.9   | 21.7   | 21.9   | 21.8   |            | 21.7       | 21.7       |
| <b>A0A0K2FLW7</b>  | 98.6   | 100    | 98.6   | 100    | 21.7       |            | 98.7       |
| <b>A0A0K2CNL4</b>  | 97.5   | 98.9   | 97.5   | 98.9   | 21.7       | 98.9       |            |

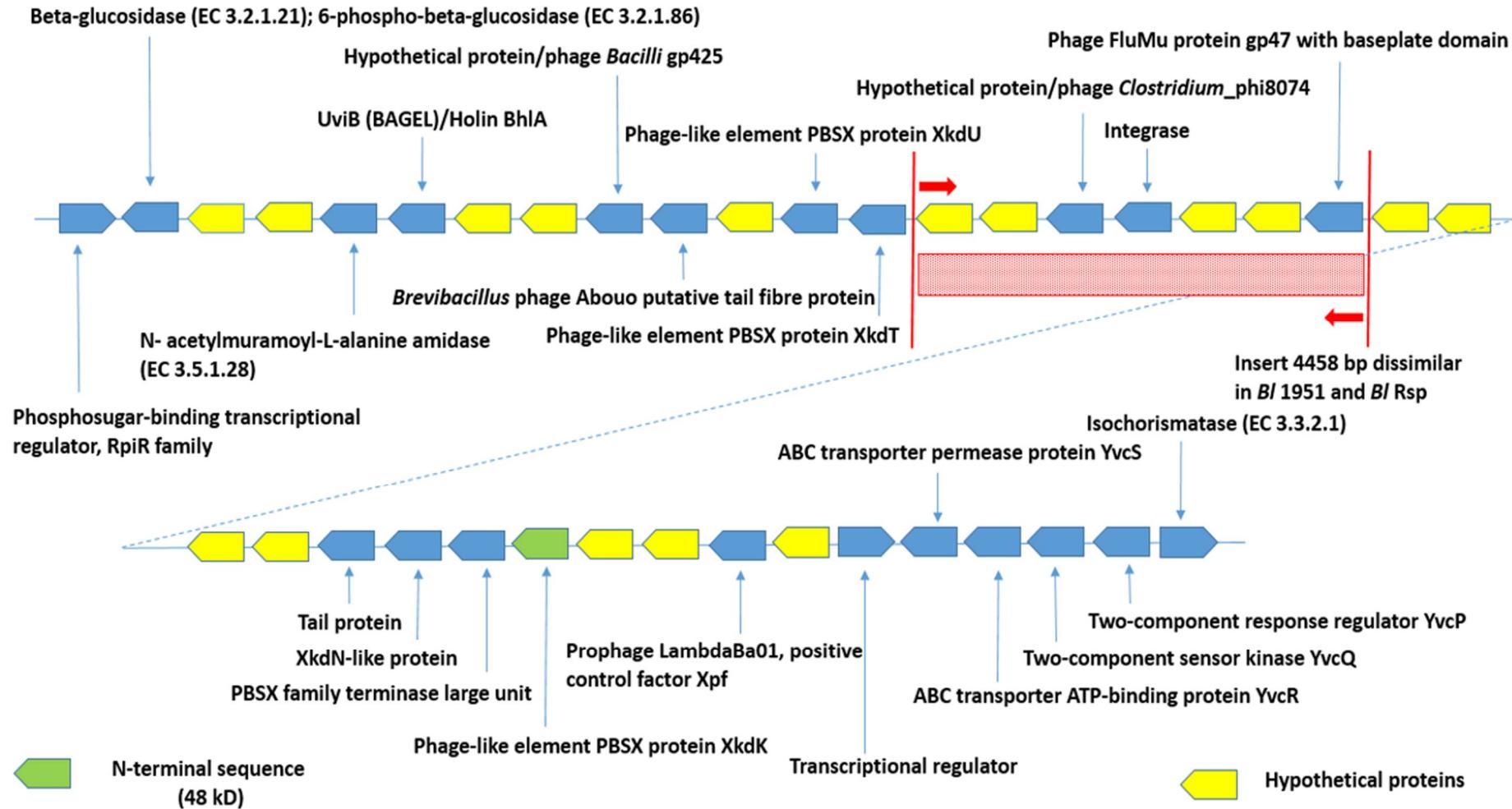


Figure S13. Schematic of the genomic architecture of *B/ 1821L* PBSX-like region encoding the identified gene *xkdK*. The red shaded box denotes *B/ 1951* region absent in *B/ 1821L*.

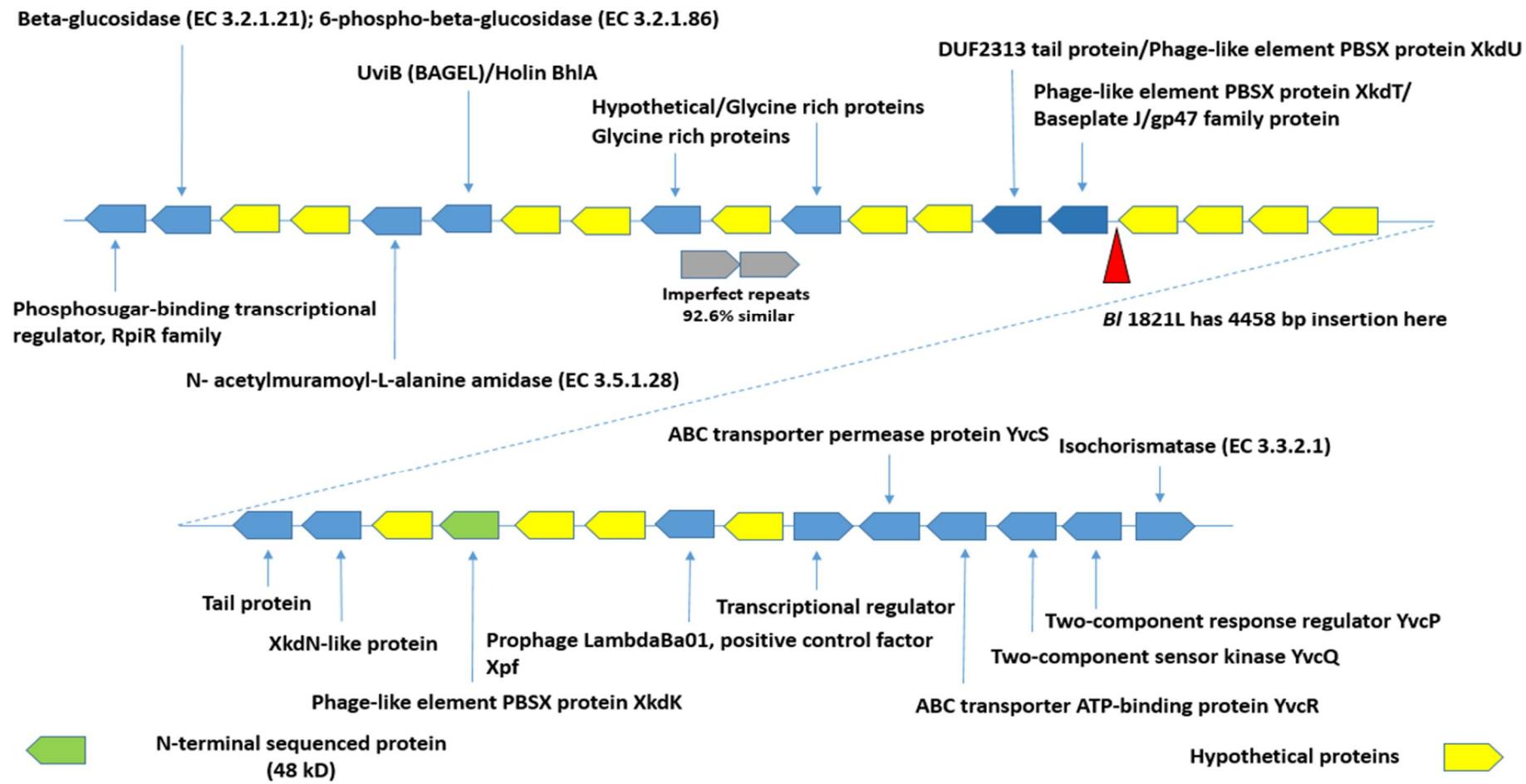


Figure S14. Genomic architecture of *B/ 1951* PBSX-like region encoding the identified gene *xkdK*. The filled red wedge points to the integration of the red shaded region of *B/ 1821L* (Figure 13) in the *B/ 1951* genome.

**Table S12. Protein orthologues of *B/1821L UviB (BAGEL)/Holin BlhA* protein in Uniprot.**

| Accession  | Homologous protein      | Organism                                       | % identity |
|------------|-------------------------|--|------------|
| AOA075R9K7 | Uncharacterised protein | <i>Brevibacillus laterosporus</i><br>LMG 15441 | 100        |
| S5M5U3     | Bacteriocin UviB        | <i>Brevibacillus</i> phage Emery               | 92.0       |
| S5M6A1     | Uncharacterised protein | <i>Brevibacillus</i> phage Davies              | 90.8       |
| S5MNE1     | Uncharacterised protein | <i>Brevibacillus</i> phage Jimmer 1            | 90.8       |
| AOA0K2CND1 | Uncharacterised protein | <i>Brevibacillus</i> phage Osiris 1            | 90.8       |

**Figure S15. Amino acids alignment of identified *B*/1821L and *B*/1951 phage-like element PBSX protein XkdK (A0A518VEB0) with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180 using programme CLUSTALO.**

|            |                    |     |  |     |
|------------|--------------------|-----|--|-----|
| P54331     | XKD <b>K</b> BACSU | 1   | MNGGTFTTGKEKERAGIYFNFKTTAQERVSLSERGTVALPVASSWGEAKTFVSISSVEDL                           | 60  |
| A0A518VEB0 | A0A518VEB0_BRELA   | 1   | -----MTIQRERPGVNVELAKAKAQERV-LPKSGVVLVPYLAENGAPDQVITMKGYEER                            | 52  |
| R4JQA6     | R4JQA6_9CAUD       | 1   | MNGGTFTPGTEKKRPGIYFNFKTTAEQRITLGERGTVALPLVMSWGEPKTFISVSDMEDL                           | 60  |
| E0U1S9     | E0U1S9_BACPZ       | 1   | MNGGTFTTGKEKERAGIYFNFKTTAQERVSLGERGTVALPVASSWGEAKTFVSISSVEDL                           | 60  |
|            |                    |     | : : * * : . : : * . * : * ; * : * . * . : * . : * :                                    |     |
| P54331     | XKD <b>K</b> BACSU | 61  | NKKVGLSIDDP <i>S</i> LLLREAKKNAKTVL <b>M</b> YRLTEGVR--ASADIAEGVKATAVYGGTKGND          | 118 |
| A0A518VEB0 | A0A518VEB0_BRELA   | 53  | V---AETFGH--IGILELPAEGGAIVLGYRMTSGNGVAASYTQSDFTLEARYPGLVGND                            | 107 |
| R4JQA6     | R4JQA6_9CAUD       | 61  | NKKVGLNIDDK <i>S</i> LLLREAKKKAQIVL <b>L</b> YRLNEGEP--AKAEIAENFVVTANYGGQKGNE          | 118 |
| E0U1S9     | E0U1S9_BACPZ       | 61  | NKKVGLSIDDP <i>S</i> LLLREAKKNAKTVL <b>M</b> YRLTEGVR--ASADIAEGVKATAVYGGTKGND          | 118 |
|            |                    |     | . . . . : : . * : . *** * * : . * . : . . * * * * : .                                  |     |
| P54331     | XKD <b>K</b> BACSU | 119 | IIIRINQNVLDAASFDTVTTYMDESEVD <b>K</b> QTVKKAEELT---ANGYVTFITGTGDLSSSTIP                | 174 |
| A0A518VEB0 | A0A518VEB0_BRELA   | 108 | IQISIKESTVELGKKELQVK-GPIKTEKFESFANVDELVTKAEQSIYIKVKKLGDKVA---                          | 163 |
| R4JQA6     | R4JQA6_9CAUD       | 119 | ITIQAENVLDSTKRDVITYL <b>G</b> DIVDKQVVKDVKDLV---KNKYVQFSGEGEAVI---                     | 171 |
| E0U1S9     | E0U1S9_BACPZ       | 119 | IIIRINPNVLDAASFDTVTTYMDESEVD <b>K</b> QTVKKAEELT---ANGYVTFITGTGDLSSSTIP                | 174 |
|            |                    |     | : * : . . : . : . . . : * . . . : . . * : . . * : . . * : .                            |     |
| P54331     | XKD <b>K</b> BACSU | 175 | LTGSEGDTAAETLN <i>S</i> AGIRLSGGTD <b>K</b> APV--NSDYDFLAAETESFDVIALPVAEGDQ            | 232 |
| A0A518VEB0 | A0A518VEB0_BRELA   | 164 | -----EETMMISLEG <b>T</b> SGITSLTANDFTTLFNSISGTD <b>F</b> DAMYLPSADVAI                  | 210 |
| R4JQA6     | R4JQA6_9CAUD       | 172 | -----TAGAA <i>S</i> GGKNGVAS--VADYTAFL <b>E</b> AAETEYFDVIALPVDNSEQ                    | 214 |
| E0U1S9     | E0U1S9_BACPZ       | 175 | LTGSEGDTAAETLN <i>S</i> AGIRLSGGTD <b>K</b> APV--NSDYDFLAAETENFDVIVLPVAEGDQ            | 232 |
|            |                    |     | : . * . * . . . * : * : : . . . * : . . * : . . * : . . * : .                          |     |
| P54331     | XKD <b>K</b> BACSU | 233 | LKAT-----AAFI <b>K</b> RIRD <b>G</b> Q <b>Q</b> KVOGV <b>T</b> ANYAGDYEGIINVTEGVILLEDG | 278 |
| A0A518VEB0 | A0A518VEB0_BRELA   | 211 | QAAAKQEMSDRELFSKKRS <b>T</b> LVTGGI <b>K</b> DKDNMNEHVQRSVANNSSRRVVNCIAAGQHVNG         | 270 |
| R4JQA6     | R4JQA6_9CAUD       | 215 | LKAT-----ASF <b>T</b> ERLRDKQGRKV <b>Q</b> GV <b>V</b> ANYADQEGIINVTS <b>G</b> VILLEDG | 260 |
| E0U1S9     | E0U1S9_BACPZ       | 233 | LKAT-----AAFI <b>K</b> RIRD <b>G</b> Q <b>Q</b> KVOGV <b>T</b> ANYAGDYEGIINVTEGVILLEDG | 278 |
|            |                    |     | * : * . . * * : . . : * . . * . * : . . : * : . . * : .                                |     |

|            |                  |     |   |     |
|------------|------------------|-----|---|-----|
| P54331     | XKDK_BACSU       | 279 | TEVTPDKATAWVAGASAGATFNQSLT--FVEYEGAVDVLHRLDHDTIVERLGKGEFLFTF    | 336 |
| A0A518VEB0 | A0A518VEB0_BRELA | 271 | KTYGSLEWAANLAGMIAATPAHISLSAQVLPLKKA---EKDWGHTDIQAAINSGTLIAVR    | 327 |
| R4JQA6     | R4JQA6_9CAUD     | 261 | TELTPAOTTAWVAGASAGANFNQSLT--FVEYEGAVDTLERLDNDQVEYRLSQGEFLFTF    | 318 |
| EOU1S9     | EOU1S9_BACPZ     | 279 | TEVTPDKATAWVAGASAGATFNQSLT--FVEYEGAVDVLHRLDHDTIVERLGKGEFLFTF    | 336 |
|            |                  |     | .   | .   |
| P54331     | XKDK_BACSU       | 337 | DARDKSVSVEKDINSIVTFTAEKNKKFAAKNKIVRVLDLAVNNNDLTRELKALIKSRKGSGSD | 396 |
| A0A518VEB0 | A0A518VEB0_BRELA | 328 | D--GDVYLIESAVNLTITLKAAREDFGKIRVSMTLDQIVVNDIISVGKKYKGK--LDNNND   | 383 |
| R4JQA6     | R4JQA6_9CAUD     | 319 | DARDRTVSVEKDINSLTSFTVEKNOQMAKNKIIIRVLDAINNDLTRELKALIKLRKANGND   | 378 |
| EOU1S9     | EOU1S9_BACPZ     | 337 | DARDKSVSVEKDINSIVTFTAEKNKKFAAKNKIVRVLDLAVNNNDLTRELKALIKSRKGSGSD | 396 |
|            |                  |     | *   | ... |
| P54331     | XKDK_BACSU       | 397 | IPASEDGLQYVKTMITQYMTTLQDAGGT-T-GEDSDEDITISMNEDRDGFLIDLAVQPVDA   | 455 |
| A0A518VEB0 | A0A518VEB0_BRELA | 384 | -----GGATFVG-AVKTYLEVREAQGAI DQGWI-FEDKKNGI-GDKRG--FRLAAKPLDA   | 434 |
| R4JQA6     | R4JQA6_9CAUD     | 379 | IPASDDGVQLVKTLITQYLTQLQDGSGIT-T-GFNSETDIVIGLNEDRDGFIIDLAVQPVDA  | 437 |
| EOU1S9     | EOU1S9_BACPZ     | 397 | IPASEDGLQYVKTMITQYMTTLQDAGGT-T-GEDSDEDITISMNEDRDGFLIDLAVQPVDA   | 455 |
|            |                  |     | *   | ... |
| P54331     | XKDK_BACSU       | 456 | AEKFYFNVEVN   | 466 |
| A0A518VEB0 | A0A518VEB0_BRELA | 435 | IIFDIEWEVL  | 445 |
| R4JQA6     | R4JQA6_9CAUD     | 438 | AEKFYFNVEVK   | 448 |
| EOU1S9     | EOU1S9_BACPZ     | 456 | AEKFYFNVEVN   | 466 |
|            |                  |     | *   | ... |

\*Highlighted dark grey color amino acids denote similarity among the aligned phage-like element PBSX protein XkdK.  
 (Refer to SM Tables S13 & S14)

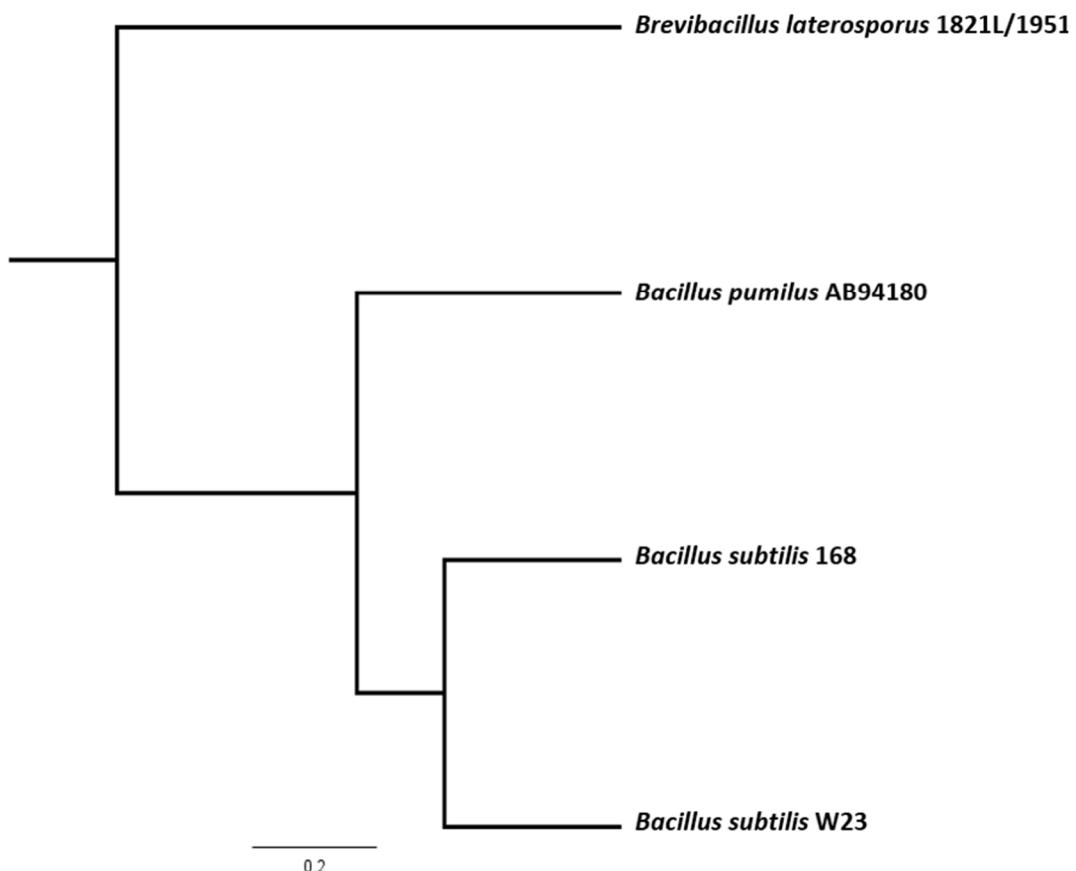
**Table S13.** Distance matrices of identified *B*/1821L and *B*/1951 phage-like element PBSX protein XkdK (A0A518VEB0) with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180.

| Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |            |        |        |
|--|--|------------|--------|--------|
|  | P54331   | A0A518VEB0 | R4JQA6 | E0U1S9 |
| <b>P54331</b>  |  | 1.45       | 0.37   | 0.01   |
| <b>A0A518VEB0</b>  | 1.45   |            | 1.40   | 1.47   |
| <b>R4JQA6</b>  | 0.37   | 1.40       |        | 0.37   |
| <b>E0U1S9</b>  | 0.01   | 1.47       | 0.37   |        |

(Refer to SM Figure S15)

**Table S14.** Amino acids alignment (%) of identified *Bi* 1821L and *Bi* 1951 phage-like element PBSX protein XkdK (A0A518VEB0) with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180.

| Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |            |        |        |
|--|--|------------|--------|--------|
|  | P54331   | A0A518VEB0 | R4JQA6 | E0U1S9 |
| <b>P54331</b>  |  | 22.9       | 66.5   | 98.7   |
| <b>A0A518VEB0</b>  | 22.9   |            | 21.6   | 22.9   |
| <b>R4JQA6</b>  | 66.5   | 21.6       |        | 66.5   |
| <b>E0U1S9</b>  | 98.7   | 22.9       | 66.5   |        |



**Figure S16.** Dendrogram showing alignment of the identified phage-like element PBSX protein XkdK (A0A518VEB0) of *Bl* 1821L and *Bl* 1951 with similar proteins of defective prophages of *Bs* W23 (E0U1S9), *Bs* 168 (P54331), and *Bacillus* phage PBP180 (R4JQA6). Key is 0.2 nt substitutions per site.

**Figure S17. Amino acids alignment of tail protein (AOA518VEA0) encoded in PBSX-like region of *B*/1821L and *B*/1951 with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180 using programme CLUSTALO.**

|            |                  |     |   |     |
|------------|------------------|-----|---|-----|
| AOA518VEA0 | AOA518VEA0_BRELA | 1   | -----MNNDMVVELLSVQKEMYKVRQGMMWERKDA--VSL--QVAWGQMGRDFLRE      | 47  |
| EOU1T3     | EOU1T3_BACPZ     | 1   | MAKLTARFDMEDRVS-----KKLRKVQNGFRALEKYRKVMQRKSAIDIRKDSKILRT     | 53  |
| R4JJ03     | R4JJ03_9CAUD     | 1   | -----   | 0   |
| P54334     | XKDO_BACSU       | 1   | MAKLTARFDLEDKVS-----KKLKRIHKGFQEVEKKVKTINRQIKISIRAEDQAFY--    | 51  |
|            |                  |     |   |     |
| AOA518VEA0 | AOA518VEA0_BRELA | 48  | TDKMERRIK-----ELHSQLKRLGT-----                                | 67  |
| EOU1T3     | EOU1T3_BACPZ     | 54  | IDRIQKSLKKKLGSQMISISAEDNASAVIQQVQTQLQGLPASVSINIGASDQATERFERL  | 113 |
| R4JJ03     | R4JJ03_9CAUD     | 1   | -----   | 0   |
| P54334     | XKDO_BACSU       | 52  | -----   | 51  |
|            |                  |     |   |     |
| AOA518VEA0 | AOA518VEA0_BRELA | 68  | -----   | 67  |
| EOU1T3     | EOU1T3_BACPZ     | 114 | RELVAGFKGFTIVLSAEDQVSPAVQKIQRYMETALKNGYSVTIRVIDHVMKTVGRISAGI  | 173 |
| R4JJ03     | R4JJ03_9CAUD     | 1   | -----   | 0   |
| P54334     | XKDO_BACSU       | 52  | -----   | 51  |
|            |                  |     |   |     |
| AOA518VEA0 | AOA518VEA0_BRELA | 68  | ---SVQAKLRVTIDDQATQKISQ-----MRSQISQSLVISGGGSSGGSKASVMDPMG     | 116 |
| EOU1T3     | EOU1T3_BACPZ     | 174 | DALTGKDNLKLELAINDKVSDKLDSLQKRIDSMSRGSPDKAAPSAAGGNTGDIASMFDPET | 233 |
| R4JJ03     | R4JJ03_9CAUD     | 1   | -----   | 0   |
| P54334     | XKDO_BACSU       | 52  | -----   | 51  |
|            |                  |     |   |     |
| AOA518VEA0 | AOA518VEA0_BRELA | 117 | DY-----LINSYNDIAPDSQAAAKE-----RGLFMARGKT---ELEGQ---ELD        | 154 |
| EOU1T3     | EOU1T3_BACPZ     | 234 | ILTALDKFASSFMEKVDEIATKFSPETILTELDKFTTSFMNKVDIAITKFSPETILTQLD  | 293 |
| R4JJ03     | R4JJ03_9CAUD     | 1   | -----   | 0   |
| P54334     | XKDO_BACSU       | 52  | -----   | 51  |
|            |                  |     |   |     |
| AOA518VEA0 | AOA518VEA0_BRELA | 155 | -----RSVGRMTQINPEM  | 167 |
| EOU1T3     | EOU1T3_BACPZ     | 294 | KFTTSFMSKVDIAITKFSPETILTQLDKFTTSFMSKVDIAITKFSPETILGQLDKFTTSF  | 353 |
| R4JJ03     | R4JJ03_9CAUD     | 1   | -----   | 0   |
| P54334     | XKDO_BACSU       | 52  | -----   | 51  |

|            |            |        |     |   |     |
|------------|------------|--------|-----|---|-----|
| A0A518VEA0 | A0A518VEA0 | BRELA  | 168 | -----MKAEEATAIYNRSDEVNPTN----KAEYAEFAAKLSMSTGFTSDQSLKMMALL    | 215 |
| E0U1T3     | E0U1T3     | BACPZ  | 354 | MSKVDAIATKFSPETILEOLDKFTTSFMGVDAIAAKFSPET-ILSQL-DKFTTSFMNKV   | 411 |
| R4JJ03     | R4JJ03     | _9CAUD | 1   | -----MKISLEMDEKKLRKFNLNM-----                                 | 19  |
| P54334     | XKDO       | _BACSU | 52  | -----RLRKI-----NDYIILKFAKSLEIKVVL-DDQ---VTSKL                 | 82  |
|            |            |        |     | : : .   |     |
| A0A518VEA0 | A0A518VEA0 | BRELA  | 216 | RDSTGVNDPERLANSLQYMST-NMK-----DFSSDFVSSMIKNTSQLGLIMDTPEKMA    | 267 |
| E0U1T3     | E0U1T3     | BACPZ  | 412 | DAIATKFSPETLLKQLDKFTGSFMKKVDEIASKFSPEAIFKQLD--KFTGSEMKVDDVV   | 469 |
| R4JJ03     | R4JJ03     | _9CAUD | 20  | -----RRFSVLSMKLYQ-----GIYRDLR--SLANQLNMPKQMV                  | 52  |
| P54334     | XKDO       | _BACSU | 83  | DAIERKL-----KRLPKETKLTVSLVDRTAAYKK--IKKMS--NQKLSSELLIPDDKV    | 132 |
|            |            |        |     | : : : : .   |     |
| A0A518VEA0 | A0A518VEA0 | BRELA  | 268 | MLVGEIGN-----M---GIPSNGLPLEA-----LKDLALKMSTQGEMSKV            | 304 |
| E0U1T3     | E0U1T3     | BACPZ  | 470 | S-----KFSPEV-IFKQLDKFTGSFMKKVDDVVSKFSP-EAIFKQ                 | 507 |
| R4JJ03     | R4JJ03     | _9CAUD | 53  | ISIQAKGLDVIKSSIDRLKQSGTSPILLTFKLNDQLSGMSS-IK-----KSILQL       | 102 |
| P54334     | XKDO       | _BACSU | 133 | T-----ST-----VKRIIGYIKKN-----                                 | 146 |
|            |            |        |     | : : : : .   |     |
| A0A518VEA0 | A0A518VEA0 | BRELA  | 305 | LQRGYEADGKSPEEAKRLATIESKEVTQLLHSDNQAMGRIFMNVASIKDDNVRQE       | 364 |
| E0U1T3     | E0U1T3     | BACPZ  | 508 | LDKFADSFMKKVDDIVS-----KFSPATIFNELDKFADSFN-KK                  | 545 |
| R4JJ03     | R4JJ03     | _9CAUD | 103 | MNRRTYYMRLNMVDQATT-----AI-----QRIKKT-----                     | 127 |
| P54334     | XKDO       | _BACSU | 147 | LKNNGYTALKVIDEITK-----TV-----NR-----                          | 167 |
|            |            |        |     | : : : : .   |     |
| A0A518VEA0 | A0A518VEA0 | BRELA  | 365 | MLNEVGSGSGK---ELQYLEPLIESAGNI---SA-----G                      | 394 |
| E0U1T3     | E0U1T3     | BACPZ  | 546 | VDDVVSKFSPETIFNEELDKFTDSFMKKVDDIVSKFSPDAILTKAEDFLTNIVDKISEKFN | 605 |
| R4JJ03     | R4JJ03     | _9CAUD | 128 | -----KSLSMAKHEI-----RVSVQDNAKSKLKKRDQAES                      | 158 |
| P54334     | XKDO       | _BACSU | 168 | -----LTALRKFECTYYVKVV-----VKTKISDKINKKES                      | 198 |
|            |            |        |     | *   |     |
| A0A518VEA0 | A0A518VEA0 | BRELA  | 395 | LIVVNKVADDEA-----EKSYQAQKDQNPFYMQAQNEVRLAMTELAAATVAKDL-----   | 443 |
| E0U1T3     | E0U1T3     | BACPZ  | 606 | FINPDKI-----A-----  | 613 |
| R4JJ03     | R4JJ03     | _9CAUD | 159 | VVKEKKIKKKP---NATAATKTKENQSWLGNLGNKALKE--IEKYAGDVSDLKKEKLSP   | 213 |
| P54334     | XKDO       | _BACSU | 199 | LINKETSKKESDKSSDKGSDKNSDQKPGWVGKIK-----GWL                    | 235 |
|            |            |        |     | : : : : .   |     |
| A0A518VEA0 | A0A518VEA0 | BRELA  | 444 | -----APGFKILSNGIANVVQSFNKL-----P-QMARLAIEGVVIAFLAKKAKKSL      | 488 |
| E0U1T3     | E0U1T3     | BACPZ  | 614 | -----NKAEEKFFDNIVSKIAKKEFKSPDKIIIEK-VGEFFEKIIKGIAEKLVNLDFGGL  | 667 |
| R4JJ03     | R4JJ03     | _9CAUD | 214 | RKFWDKEKLPWETKIDEYKQEVIGRISERKIKIN-PEEY-----LDKWFNKGLDII      | 264 |
| P54334     | XKDO       | _BACSU | 236 | KDWWTSAWPWIKDILNEIGKAFLDRFKERLIEKLKNDVFDKIID-----KII          | 283 |
|            |            |        |     | : : : : .   |     |



|            |                  |      |   |      |
|------------|------------------|------|---|------|
| AOA518VEAO | AOA518VEAO_BRELA | 858  | KRIIETAFVN ALETRGGKA-----   | 876  |
| E0U1T3     | E0U1T3_BACFZ     | 958  | ATAIWSKIVNAWTTIKN VFSTV ASWFMSNVWGPVKSAVIGAATTIWDKMTGAWTKIKSV-    | 1016 |
| R4JJ03     | R4JJ03_9CAUD     | 581  | TQEKWGD GAVEIWNSITKKLGE--TVFNGEWNGEKWD SVKKWTKEKWDGAVNIWSIKGKI    | 638  |
| P54334     | XKDO_BACSU       | 590  | ATTIWTGLCNAWTVISGIWSTVSTWFMDNVNPVSEAVTTAATWIWSKINDAWTIISNI-       | 648  |
|            |                  | :    |   |      |
| AOA518VEAO | AOA518VEAO_BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ     | 1017 | FSTVSGWFMDIVWNPVKNTVLDVGKG ISDAFKKAIDTVKNIWKGLSG                  | 1063 |
| R4JJ03     | R4JJ03_9CAUD     | 639  | RET VFNKD WGEKWD DVKNWSE-NKWEQSKTIWS-AAKATASSTLFNKNWWTNKWNDVKS    | 696  |
| P54334     | XKDO_BACSU       | 649  | -----WTGVSNWFYDNVWNPIVKKVEDIKTSIKEKFDDARKFFEDAWDGIET              | 695  |
| AOA518VEAO | AOA518VEAO_BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ     | 1064 | WFKKNIQEPLTKVGEAISDAFSKA FGWVKQIWDKAGGVASKVINFVTGGGD-----         | 1114 |
| R4JJ03     | R4JJ03_9CAUD     | 697  | WGKNILG-----DTWDLIKSKGAEVIGRH-----IV--                            | 722  |
| P54334     | XKDO_BACSU       | 696  | WFNEHVKDPLVKVSEKIAEK---FQWLFDLKDSVGGFVAKVIHRGEEVTHLKKKPIKKD       | 751  |
| AOA518VEAO | AOA518VEAO_BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ     | 1115 | -----PNKGKD PDKKATGGYITKPTISWIGEAGKEFVIPV                         | 1149 |
| R4JJ03     | R4JJ03_9CAUD     | 723  | -----KFEKGREKGRKDFKPDQKATGGYITQPTLSWIGEAGNEFVIPT                  | 765  |
| P54334     | XKDO_BACSU       | 752  | SKEKKTSSKPTTNNIFAQSPSNNNMTVFQQALDQNATGGYITKPTISWIGEAGKEFVIPV      | 811  |
| AOA518VEAO | AOA518VEAO_BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ     | 1150 | DNNRGRGKMLLSQAASKLGMQVVDDMG-AASSSSGNPVSVSGGAASSPLSGSASP-SMDT      | 1207 |
| R4JJ03     | R4JJ03_9CAUD     | 766  | QNNRGRGKMLLAQAA SHLGMSSVPPSSGAASPVSSSPAPITPNAPAAPTASAGSIGGTVS     | 825  |
| P54334     | XKDO_BACSU       | 812  | DNNRGRGKMLLSQAASKLGMQVVDDMG-AASSSSGSPASVSGGAAVSPLSGTASP-VMNT      | 869  |
| AOA518VEAO | AOA518VEAO_BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ     | 1208 | ANLTGQASTLGQQFSEGFGKG ISNQPVKMEDWKKNINTPFTQMISSSQNYGKQMVG YA      | 1267 |
| R4JJ03     | R4JJ03_9CAUD     | 826  | MNGNIQSASIGE QFNHD FEQGLNQKVVS LDQWKQKN IQQPFNQMTSDSGKYGQQT VSAFA | 885  |
| P54334     | XKDO_BACSU       | 870  | ANLTGQASTLGQQFSEGFGKG ISDQPVKMEDWKKNINTPFTQMISASPNYGKQMVG YA      | 929  |
| AOA518VEAO | AOA518VEAO_BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ     | 1268 | KGQNGTATGTDGFLQSKVKT PFQATV NKSSSWGTETVKGFAQGQN STQTGTAQYVSTHVD   | 1327 |
| R4JJ03     | R4JJ03_9CAUD     | 886  | TGQQMTPGTDSFLQSRVKA PYQVMTASPTWGS GTVSGFATGQNAT SIGHTSQYDQHVK     | 945  |
| P54334     | XKDO_BACSU       | 930  | KGQSGTATGTDGFLQSKVKT PFQATV SKSSSWGTGTVKGFAQGQN STQTGTAQYVSTHVD   | 989  |

|            |              |       |      |   |      |
|------------|--------------|-------|------|---|------|
| AOA518VEAO | AOA518VEAO   | BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ |       | 1268 | KGQNGTATGTDGLQSKVTPFQATVKSSSGTETVKGFAQGQNSTQTGTAQYVSTHVD      | 1327 |
| R4JJ03     | R4JJ03_9CAUD |       | 886  | TGQQMTPGTGDSFLQSRVKAPYQQVMTASPTIWGSGTVSGFATGQNATSIGTSQYVDQHVK | 945  |
| P54334     | XKDO_BACSU   |       | 930  | KGQSGTATGTDGLQSKVTPFQATVKSSSGTETVKGFAQGQNSTQTGTAQYVSTHVD      | 989  |
| AOA518VEAO | AOA518VEAO   | BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ |       | 1328 | KPFLRSKDTNSWGSVMGNFVTGMNSKSSEVKQAAKDMAKRVEQAFCREELDIHSPSRVM   | 1387 |
| R4JJ03     | R4JJ03_9CAUD |       | 946  | QPFLQAKQESPWGSGMIDAFNSGMRSKASEVTQAAKEMAKKVEQAFCREELDIHSPSRVM  | 1005 |
| P54334     | XKDO_BACSU   |       | 990  | KPFLRSKDTNSWGSGLIGNFVTGMNSKSSEVKQAAKDMAKRVEQAFCREELDIHSPSRVM  | 1049 |
| AOA518VEAO | AOA518VEAO   | BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ |       | 1388 | MSLGRFASIGVVKGGLGSDVVKKYAEKQAGSLAAAYS GGMAMSGNVKQWIMAALMATKPM | 1447 |
| R4JJ03     | R4JJ03_9CAUD |       | 1006 | MSLGKFASIGVVKGGLDSDVVKFAENQAGSLIGAFSGMGASGLSVQQWLMAALMATGTS   | 1065 |
| P54334     | XKDO_BACSU   |       | 1050 | MSLGRFASVGVVKGGLDSDVVKKYAEKQAGSLAAAYS GGMAGVGNVKQWIMAALMATKPL | 1109 |
| AOA518VEAO | AOA518VEAO   | BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ |       | 1448 | SWLPGLMTIAQHESGGNPNAINLWDSNAKAGHPSQGLMQTIPSTFNDHKAPSMGNIKNPI  | 1507 |
| R4JJ03     | R4JJ03_9CAUD |       | 1066 | SWLPGLMTIAQHESNGNPNAKAGHPSQGLMQTIPSTFNDHKAPSMGNIKNPI          | 1125 |
| P54334     | XKDO_BACSU   |       | 1110 | SWLSGLMTIAQYESGGNPNSINLWDSNAKAGNPSQGLMQTVPPTFNAHKAPGMGNIRNPI  | 1169 |
| AOA518VEAO | AOA518VEAO   | BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ |       | 1508 | HNAAAAAIGYIKSRYGSINNPGIKSLNHHGPYVGYYANGGLITKEQIARVGEGNKREWIIP | 1567 |
| R4JJ03     | R4JJ03_9CAUD |       | 1126 | HNAAVAINYIKGRYGVFTPGLRSMRGGPYKGYYANGGLITQEVARVGEGNKREWIIP     | 1185 |
| P54334     | XKDO_BACSU   |       | 1170 | HNAAAAAIGYIKSRYGSIDNPGIKSLRGGPYVGYYANGGLITKEQIARVGEGNKREWIIP  | 1229 |
| AOA518VEAO | AOA518VEAO   | BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ |       | 1568 | EERGIRGRYLLQKAAQALGMEVTDPSSQSQ-SDLSSGQVSAI-TAGTRQTIHTAGTKEIKI | 1625 |
| R4JJ03     | R4JJ03_9CAUD |       | 1186 | EERGIRGRYLLTQAAKALGMQVYDPNSNASAPL-PESQMQQVTSQAQSAGNTTSGNKQITI | 1244 |
| P54334     | XKDO_BACSU   |       | 1230 | EERGIRGRYLLQRAAQALGMEVTDPSSQSQSELSSGQVSAV-TSASRPTAAVSGSKEIYI  | 1288 |
| AOA518VEAO | AOA518VEAO   | BRELA | 877  | -----   | 876  |
| E0U1T3     | E0U1T3_BACFZ |       | 1626 | EFNGDQHFHNEQDADSLVAKIKQALLDELQKDINTGTVGVVAFD                  | 1669 |
| R4JJ03     | R4JJ03_9CAUD |       | 1245 | QFNGDQHFHNGQDQQLVEKIRQMLVDELEVELQTGTVGVVAFD                   | 1288 |
| P54334     | XKDO_BACSU   |       | 1289 | QFNGDQHFHNGQDAESLAAKIKQALIDELQKDINIGTGVVAFD                   | 1332 |

\*Highlighted dark grey color amino acids denote similarity among the aligned phage-like element PBSX protein XkdK.  
 (Refer to SM Tables S15 & S16)

**Table S15. Distance matrices of tail protein (A0A518VEA0) encoded in PBSX-like region of *B*/1821L and *B*/1951 with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180.**

| Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |        |        |            |
|--|--|--------|--------|------------|
|  | P54334   | E0U1T3 | R4JJ03 | A0A518VEA0 |
| <b>P54334</b>  |  | 0.56   | 0.85   | 0.71       |
| <b>E0U1T3</b>  | 0.56   |        | 0.84   | 1.73       |
| <b>R4JJ03</b>  | 0.85   | 0.84   |        | 1.79       |
| <b>A0A518VEA0</b>  | 0.71   | 1.73   | 1.79   |            |

**Table S16. Amino acids alignment (%) of tail protein (A0A518VEA0) encoded in PBSX-like region of *B*/1821L and *B*/1951 with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180.**

| Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |        |        |            |
|--|--|--------|--------|------------|
|  | P54334   | E0U1T3 | R4JJ03 | A0A518VEA0 |
| <b>P54334</b>  |  | 51     | 37     | 0          |
| <b>E0U1T3</b>  | 51.9   |        | 37.1   | 50         |
| <b>R4JJ03</b>  | 37   | 37.1   |        | 0          |
| <b>A0A518VEA0</b>  | 0  | 50     | 0      |            |

**Figure S18. Amino acids alignment of holin protein (AOA075R9K7) encoded in PBSX-like region of *B*/1821L and *B*/1951 with the similar protein of the defective prophages PBSZ, PBSX, and PBP180 using programme CLUSTALO.**

|            |                  |    |   |    |
|------------|------------------|----|---|----|
| AOA075R9K7 | AOA075R9K7_BRELA | 1  | -----MEES---VMNALQQGP-----FAAL                                  | 18 |
| E0U1U5     | E0U1U5_BACPZ     | 1  | MKMF DKGTVIRTVLLVALINQTMMLGKSPLDIQEEQVNQLADALYSAGSVIFTIGTL      | 60 |
| R4JHG4     | R4JHG4_9CAUD     | 1  | MKTFDKGTVIRTVLLFIALINQTLV MFG QTVLP ISEEQVQTAGEALYVAGSTIFTMVTAV | 60 |
| Q99163     | XHLB_BACSU       | 1  | MNTFDKGTVIRTVLLIALINQTMMLGKSPLDIQEEQVNQLADALYSAGSIAFTIGTL       | 60 |
|            |                  |    | ***. :** * ***  |    |
| AOA075R9K7 | AOA075R9K7_BRELA | 19 | FVWL LFS--TKKEGRDREALIVKQAQITREAKLMEHNERMVIQLERNSTLQQIERSLSGL   | 76 |
| E0U1U5     | E0U1U5_BACPZ     | 61 | A AWFKN NYVTEKGKKQRD-LIKENNLT K-----                            | 87 |
| R4JHG4     | R4JHG4_9CAUD     | 61 | I AWFKN NYVTEKGQLQKD-ALKQRGLTK-----                             | 87 |
| Q99163     | XHLB_BACSU       | 61 | A AWFKN NYVTEKGKKQRD-LIRDNNLT K-----                            | 87 |
|            |                  |    | . *: . * * ::: * .. *:  |    |
| AOA075R9K7 | AOA075R9K7_BRELA | 77 | EMELQELKEKVE  | 88 |
| E0U1U5     | E0U1U5_BACPZ     | 88 | -----   | 87 |
| R4JHG4     | R4JHG4_9CAUD     | 88 | -----   | 87 |
| Q99163     | XHLB_BACSU       | 88 | -----   | 87 |

\*Highlighted dark grey color amino acids denote similarity among the aligned holin proteins.  
 (Refer to SM Figure S19 & Tables S17 and S18)

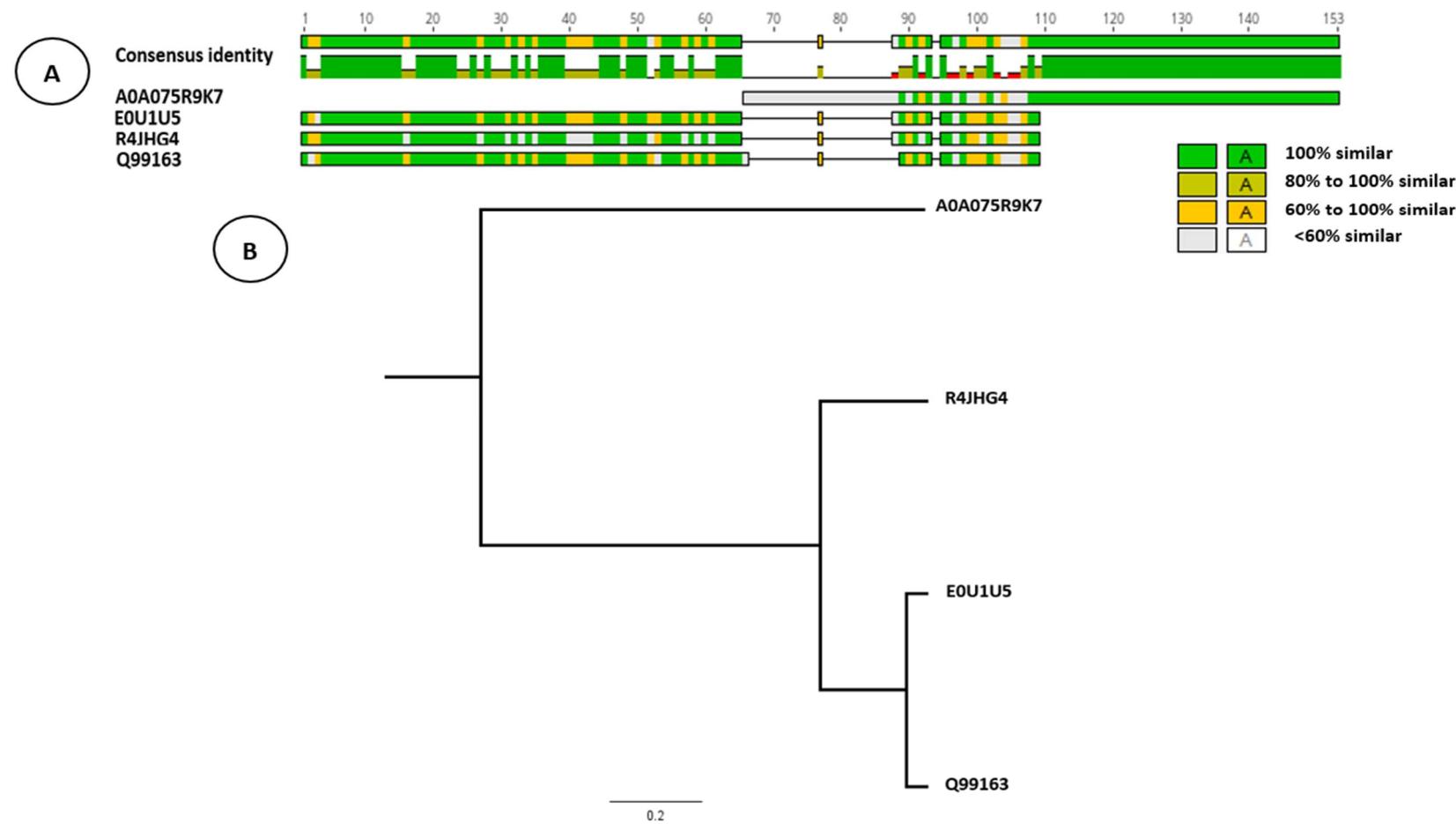
**Table S17.** Distance matrices of holin protein (A0A075R9K7) encoded in PBSX-like region of *Bl* 1821L and *Bl* 1951 with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180.

| Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |        |        |        |
|--|--|--------|--------|--------|
|  | A0A075R9K77  | E0U1U5 | R4JHG4 | Q99163 |
| <b>A0A075R9K77</b>   |  | 2.41   | 1.48   | 1.89   |
| <b>E0U1U5</b>  | 2.41   |        | 0.45   | 0.08   |
| <b>R4JHG4</b>  | 1.48   | 0.45   |        | 0.47   |
| <b>Q99163</b>  | 1.89   | 0.08   | 0.47   |        |

**Table S18.** Amino acids alignment (%) of holin protein (A0A075R9K7) encoded in PBSX-like region of *Bl* 1821L and *Bl* 1951 with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180.

| Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |        |        |        |
|--|--|--------|--------|--------|
|  | A0A075R9K77  | E0U1U5 | R4JHG4 | Q99163 |
| <b>A0A075R9K77</b>   |  | 13.6   | 11.4   | 13.6   |
| <b>E0U1U5</b>  | 13.6   |        | 64.4   | 89.8   |
| <b>R4JHG4</b>  | 11.4   | 64.4   |        | 61.4   |
| <b>Q99163</b>  | 13.6   | 89.8   | 61.6   |        |

**Figure S19. Schematic presentation of amino acids alignment of holin protein (A0A075R9K7) encoded in PBSX-like region of *Bi* 1821L and *Bi* 1951 with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180 using programme Geneious basic (A) and CLUSTALO (B).**



**Figure S20.** Amino acids alignment of N-acetylmuramoyl-L-alanine amidase protein (A0A518VEA4) encoded in PBSX-like region of *B. 1821L* and *B. 1951* genome with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180 using the programme CLUSTALO.

|            |                  |     |   |     |
|------------|------------------|-----|---|-----|
| AOA518VEA4 | AOA518VEA4_BRELA | 1   | -----MEKPLIIIDAGHG---GADPGASGNHMQKEKDQLQIGLYQ                 | 37  |
| EOU1U7     | EOU1U7_BACFZ     | 1   | MVNIIQDFIPVGANNRPGYAMTPLYITVHNNTANTAAAGADAEEAHARYLKNPDTATS    | 55  |
| R4JMV0     | R4JMV0_9CAUD     | 1   | MVKIIQALIPKQNRRNPGNRMKPLYITVHNNTNTGRGADAANHAFAVARASTGVS       | 55  |
| P39800     | XLYA_BACSU       | 1   | MVNIIQDFIPVGANNRPGYAMTPLYITVHNNTANTAVGADAAAHHARYLKNPDTTS      | 55  |
|            |                  |     | * : * . . . *** . : . . .                                     |     |
| AOA518VEA4 | AOA518VEA4_BRELA | 38  | LQRCRELNLPVAITRTTDTTLTPSQRTTLVKOSGATYCISNHINAGGEGEVEAIHSI--F  | 95  |
| EOU1U7     | EOU1U7_BACFZ     | 56  | -----WHFTVDD-----TEIYQHL--PLNENGWHAGDGNNSGNRASIGIE            | 93  |
| R4JMV0     | R4JMV0_9CAUD     | 56  | -----WHYTVDD-----QVYQHL--PLNENGWHAGDGRGTGNMKSIGIE             | 93  |
| P39800     | XLYA_BACSU       | 56  | -----WHFTVDD-----TEIYQHL--PLNENGWHAGDGNNSGNRASIGIE            | 93  |
|            |                  |     | * . * : * . * : *** . * . * **                                |     |
| AOA518VEA4 | AOA518VEA4_BRELA | 96  | TSNKLANALAQAQVAAEQQRERRVYTRVGSGRDYFFMHRETGAVENTIIMEYGFIDHVGDA | 155 |
| EOU1U7     | EOU1U7_BACFZ     | 94  | ICENTDGDFAQATANAQWLTKTIMSEHN-----ISLANVVPHKYWSGKECP           | 139 |
| R4JMV0     | R4JMV0_9CAUD     | 94  | ICENAGANFEQAVKNAQWLIRKIMGDLG-----IPLSNVVPHKHWSGKECP           | 139 |
| P39800     | XLYA_BACSU       | 94  | ICENADGDFAKATANAQWLTKTMAEHN-----ISLANVVPHKYWSGKECP            | 139 |
|            |                  |     | : : : * . : : : . : : : . : : : .                             |     |
| AOA518VEA4 | AOA518VEA4_BRELA | 156 | QKLTNNKRYAEAVLKAFSSHIGHPYSPALEEPIDDFQMAVDALVQA-----           | 202 |
| EOU1U7     | EOU1U7_BACFZ     | 140 | RKLLDTWDSFKAGIGGGSGTYVVVKQGDTLTSIARAFGVTVAQLRENNNIEDPNVIQVGQ  | 199 |
| R4JMV0     | R4JMV0_9CAUD     | 140 | RKLLNRRWDGFKAGIASASTSQMTTAKPKVKEPSI-----KT                    | 174 |
| P39800     | XLYA_BACSU       | 140 | RKLLDTWDSFKAGIGGGSGTYVVVKQGDTLTSIARAFGVTVAQLQEWNNIEDPNLIRVGQ  | 199 |
|            |                  |     | * : * . : : : . : : .   |     |
| AOA518VEA4 | AOA518VEA4_BRELA | 203 | -----KIITSPDYWKQNAGFNGTVLG                                    | 223 |
| EOU1U7     | EOU1U7_BACFZ     | 200 | VLIVSAPSYYAEEPELYPLPDGIIKLTTPYTSGEHVFOVQRALAALYFPDKGAVNNNGIDG | 259 |
| R4JMV0     | R4JMV0_9CAUD     | 175 | IISTRSTSKTNVKKKIYSLPAGILKVTKPLKGSAVKAVQOALASIYFPDKGAINNGIDG   | 234 |
| P39800     | XLYA_BACSU       | 200 | VLIVSAPSAAEKPELYPLPDGIIQLTTPYTSGEHVFOVQRALAALYFPDKGAVNNNGIDG  | 259 |
|            |                  |     | : : : : : : : * : *   |     |
| AOA518VEA4 | AOA518VEA4_BRELA | 224 | DYAAQLIKNMA-----NHLKAGA-----                                  | 241 |
| EOU1U7     | EOU1U7_BACFZ     | 260 | IYGPKTADAVARFQSUNGITADGIYGPATKAKIAAQLS-                       | 297 |
| R4JMV0     | R4JMV0_9CAUD     | 235 | YIGPKTANAVSRFQMMGHTPDGIYGPKTKETLSKVIES                        | 273 |
| P39800     | XLYA_BACSU       | 260 | VYGPKTADAVARFQSUNGITADGIYGPATKEKIAAQLS-                       | 297 |
|            |                  |     | * : * . : : : . : : .   |     |

\*Highlighted dark grey color amino acids denote similarity among the aligned N-acetylmuramoyl-L-alanine amidase proteins.  
(Refer to SM Figure S21 & Tables S19 and S20)

**Table S19.** Distance matrices of N-acetylmuramoyl-L-alanine amidase protein (A0A518VEA4) encoded in PBSX-like region of *B*/*I* 1821L and *B*/*I* 1951 with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180 .

| Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |        |        |        |
|--|--|--------|--------|--------|
|  | A0A518VEA4   | E0U1U7 | R4JMV0 | P39800 |
| <b>A0A518VEA4</b>  | 0.22   | 1.75   | 1.40   |        |
| <b>E0U1U7</b>  | 0.22   | 0.55   | 0.05   |        |
| <b>R4JMV0</b>  | 1.75   | 0.55   | 0.54   |        |
| <b>P39800</b>  | 1.40   | 0.05   | 0.54   |        |

**Table S20.** Alignment (%) of N-acetylmuramoyl-L-alanine amidase protein (A0A518VEA4) encoded in PBSX-like region of *B*/*I* 1821L and *B*/*I* 1951 with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180.

| Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) | Uniprot Accession #<br>( <a href="https://www.uniprot.org">https://www.uniprot.org</a> ) |        |        |        |
|--|--|--------|--------|--------|
|  | A0A518VEA4   | E0U1U7 | R4JMV0 | P39800 |
| <b>A0A518VEA4</b>  | 15.4   | 7.7    | 15.4   |        |
| <b>E0U1U7</b>  | 15.4   | 53.5   | 95.3   |        |
| <b>R4JMV0</b>  | 7.7  | 53.5   | 53.9   |        |
| <b>P39800</b>  | 15.4   | 95.3   | 53.9   |        |

**Figure S21. Schematic presentation of amino acids alignment of N-acetyl muramoyl-L-alanine amidase protein (A0A518VEA4) encoded in PBSX-like region of *B1* 1821L and *B1* 1951 with the similar proteins of the defective prophages PBSZ, PBSX, and PBP180 using programme Geneious basic (A) and CLUSTALO (B).**

