Isolation and molecular characterisation of *Achromobacter* phage phiAxp-3, an N4-like bacteriophage

Yanyan Ma[1#], Erna Li[2#], Zhizhen Qi[3#], Huan Li[4], Xiao Wei[4], Weishi Lin[4], Ruixiang Zhao[1], Aimin Jiang[2], Huiying Yang[5], Zhe Yin[5*], Jing Yuan[4*], Xiangna Zhao[4*]

[1]College of Food Science, Henan Institute of Science and Technology, Xinxiang, China, 453003

[2]College of Food Science, South China Agricultural University, Guangzhou, China , 510642

[3]Qinghai Institute for Endemic Disease Prevention and Control of Qinghai Province, Xining, China, 810000

[4]Institute of Disease Control and Prevention, Academy of Military Medical Sciences, Beijing, China, 100071

[5]State Key Laboratory of Pathogen and Biosecurity, Beijing Institute of Microbiology and Epidemiology, Beijing, China, 100071

[#]: These authors contributed equally to this work.

*Correspondence: Zhe Yin, State Key Laboratory of Pathogen and Biosecurity, Beijing Institute of Microbiology and Epidemiology, Beijing, China; Jing Yuan, Institute of Disease Control and Prevention, Academy of Military Medical Sciences, Beijing, China; Xiangna Zhao, Institute of Disease Control and Prevention, Academy of Military Medical Sciences, Beijing, China
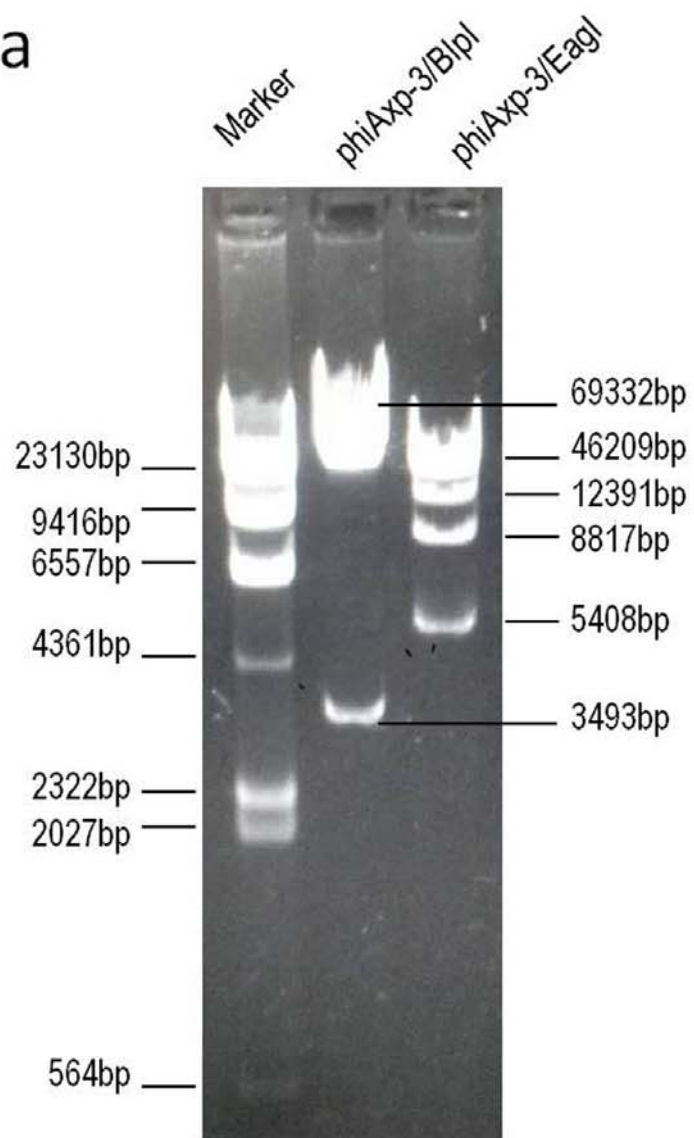
Tel: +86-10-66948434; Fax: +86-10-66948301

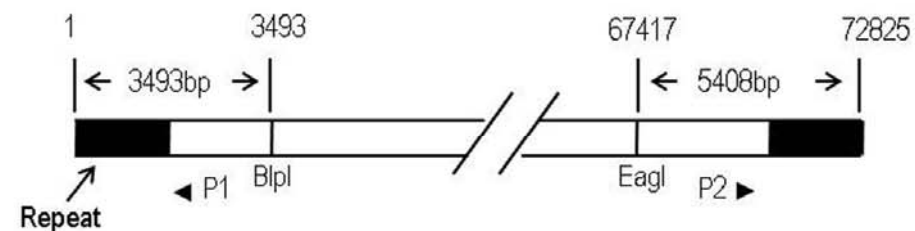E-mail: jerry9yin@163.com; yuanjing6216@163.com; xnazhao@163.com

Supplementary Figure legends

Supplementary Figure 1. Schematic illustration of the structure and linearity of phiAxp-3 genomic ends. (a) Restriction fragment length polymorphism analysis of phiAxp-3 DNA. Genomic DNA from phage phiAxp-3 was digested with the enzymes indicated (*Blp*I and *Eag*I) and run on an agarose gel (0.7%). (b) Sequence analysis of the ends of the phiAxp-3 genome. Black arrows indicate the oligonucleotide primers (P1 and P2) that were used, and the line with two arrowheads indicates the end fragments produced by genome digestion with *Blp*I and *Eag*I restriction enzymes. Genome numbering of the sequences is that of the revised sequence. The repeat regions are shown in black at both ends of the genome. (c) Diagram showing the arrangement of the phiAxp-3 genome as originally sequenced and the revised genome arrangement.

Supplementary Figure 2. Multiple sequence alignment of the direct terminal repeats of phage phiAxp-3 with JWAlpha and JWDelta. The alignment was performed using ClustalX. Positions with a single, fully conserved base pair are indicated by an asterisk (*).

a

Marker　phiAxp-3/BlpI　phiAxp-3/EagI

23130bp
9416bp
6557bp
4361bp
2322bp
2027bp
564bp

69332bp
46209bp
12391bp
8817bp
5408bp
3493bp

b

End sequencing of phiAxp-3 genomic DNA

1　　　　3493　　　　　　67417　　　72825

← 3493bp →　　　　　　← 5408bp →

Repeat　　◄ P1　　BlpI　　　　　EagI　　P2 ►

P1: TGATCGACATGGTAATACTCC
P2: ACTACTACAAGAACATTAC

c

1　413　　　　　　　　　　72406　72409

Originally genome

1 3　416　　　　　　　72409　72412　72825

Revised genome
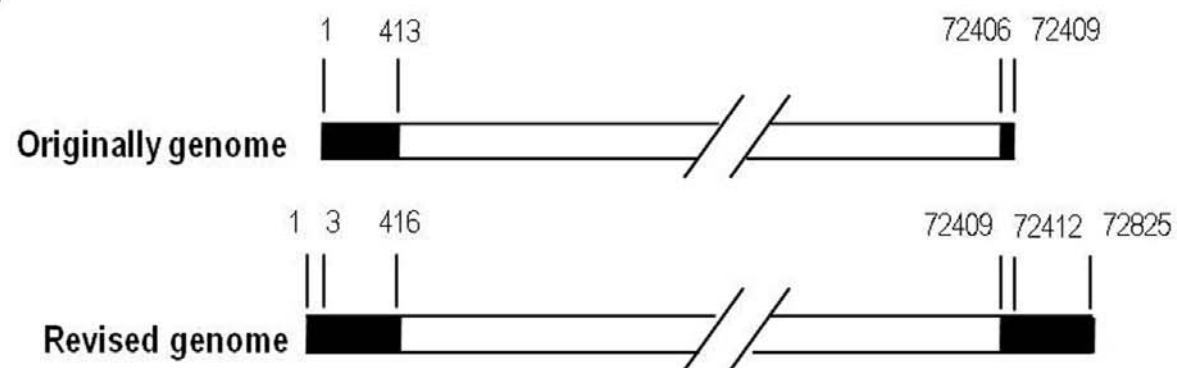
```
JWAlpha    ------------------------------ACACACCCCCCCGGTGTCTTGCTCCTGTGCAC
JWDelta    --------------------------------------CCCCCGGTGTCTTGCTCCTGTGCAC
phiAxp-3   TATGTGTACACGGTACACCCTACACTTTATACACCCCCCCGGGGTCTCTC-CTAGCCCAC
                                               ******* ****   *  *    *   ***

JWAlpha    CTCTACCCA--CCCCCATGTTGTTGACGCTCCGCGTCTTGTGGGTCTGTTGCCTATCATC
JWDelta    CTCTACCCA--CCCCCATGTTGTTGACGCTCCGCGTCTTGTGGGTCTGTTGCCTATCATC
phiAxp-3   ACCTACCCTACCCCCCATGTTTCTGACGCTCCGCGTCTTCTGGGTATGTTGCC---CATC
            ******    ** ********** ***************** ***** *******   ****

JWAlpha    TATCAATCTTCTTAGGAGTATTAAC-ATGTCCACTTCCACCGCTCGCATGACCATCGGCT
JWDelta    TATCAATCTTCTTAGGAGCATTAAC-ATGTCCACTTCCACCGCTCGCATGACCATCGGCT
phiAxp-3   TATC--TTTCTTAGGAGTATCAACTATGTCCACGTCCACCGCTCGCCTGACCATCGGCT
            ****   * *  ******* ** *** ******* ************* *********** ***********

JWAlpha    CTGCTCTCGGTACGATCAATACCGCTGCGCTTACCATCTCGTCCACTCTGGATGCAGTTG
JWDelta    CTGCTCTCGGTACGATCAATACCGCTGCGCTTACCATCTCTTCCACTCTGGATGCAGTTG
phiAxp-3   CTCTGCTCGGTACCGTGAATACTGCCGCCATCACCGTGTCGTCCACTCTGGATGCTGCTG
            **     *******  *  ***** **  **   * ***  *  ** ************* * **

JWAlpha    GCTCTGGTGTTGGTATGCTCAACGCCATGGTTGACAAGGCTGCTCGTGAACAACGTATGC
JWDelta    GCTCTGGTGTTGGTATGCTCAACGCCATGGTTGACAAGGCTGCTCGTGAACAACGTATGC
phiAxp-3   GTGCTGCTGTCGGTATGCTCAACTCTGCTGTCTCCAAGGCAGCTAAAGAGCAAGCTATCC
            *   *** *** ************ *      **  ****** ***   ** *** *** *

JWAlpha    GTCACAAAGCTGACGTCTCCGAGTTCAAGCACCGTCTCATCAACGAAGTAGCTATGGCTC
JWDelta    GTCACAAAGCTGACGTCTCCGAGTTCAAGCACCGTCTCATCAACGAAGTAGCTATGGCTC
phiAxp-3   GTCACAAGGTTGACGTCAACGAGTTCAAGAGTCGCCTGATCATGGAAGTCTCCATGGCTC
            ******* * *******  ************   ** ** ****   *****   * *******

JWAlpha    GTGCACAACGTGAACGTCAAGTCATAGAGTTCTGCA------------------------
JWDelta    GTGCACAACGTGAACGTCAAGTCATAGAGTTCTGCAAGGACAAGGACAATGAAACCTTGT
phiAxp-3   GTGCTCAACGTGAGCGTCAAGTAGTAGAGTTCTGCAAGGACTCTGTCAACGAGAAGCTCT
            **** ******** ******* *  ***********  ***********

JWAlpha    ---------------------------------------
JWDelta    ACTCCAAGGCTCACGACGAACTCACCGAGTTGCTCAAG
phiAxp-3   TC-------------------------------------
```