

## Isolation and Identification of Two Novel Filamentous Bacteriophages in the Pandemic *Vibrio parahaemolyticus* O3:K6 and O4:K68 Strains

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*Vibrio parahaemolyticus* is a halophilic marine vibrio which causes gastroenteritis in humans by seafood consumption. Pandemic infections of *V. parahaemolyticus* serovar O3:K6 have been breaking out since 1996 in Asian countries, and the United States. The O4:K68 and O1:K untypable (O1:KUT) have been isolated from international travelers in India, Bangladesh, and Japan since 1997. All of the O3:K6, O4:K68 and O1:KUT strains isolated between 1995~1998 were shown to be genetically closely related and may be come from one clone. It is unknown why these strains have recently emerged and caused a pandemic outbreak. It has been reported that a filamentous phage, CTXf, of *Vibrio cholerae* transmits the cholera toxin gene cluster (ctxAB) horizontally from toxigenic to non-toxigenic strains. In this study, we discuss the possibility of association between the filamentous phages of *V. parahaemolyticus* and the pandemic infections.

We isolated two novel filamentous bacteriophages, designated as VfO3K6 and VfO4K68 from

the pandemic *Vibrio parahaemolyticus* strains belonging to O3:K6 and O4:K68 serovar, respectively. The genomes of the two phages were single-stranded circular DNAs, converted to the double-stranded replicative form (RF) in the host cells, and integrated into the host chromosomes. The 8,784 base pairs (bp) and 6,891 bp nucleotide sequences of the VfO3K6 and VfO4K68 RF DNAs were identical except that the VfO4K68 DNA deleted a 1,893 bp fragment presented in the VfO3K6 DNA. The VfO3K6 DNA encoded 10 putative open reading frames (ORFs) whereas the VfO4K68 DNA did 8 ORFs. Seven ORFs of the two phages were identical to each other, while the remaining ORFs were different. Nucleotide sequences homologous to the VfO3K6 and VfO4K68 DNAs were specifically existed in all of the pandemic O3:K6, O4:K68 and O1:KUT strains. Therefore, we concluded that the two filamentous phages act as horizontal genetic transmitters between the pandemic *V. parahaemolyticus* strains and might be involved in the outbreak infections caused by these emerging strains.