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NOTES

FK Phage for Differentiating the Classical and El Tor Groups of *Vibrio cholerae*

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A new vibrio-infecting phage (FK phage) isolated from sewage lysed all strains of *Vibrio cholerae* biovar *cholerae*, whereas all strains of *V. cholerae* biovar El Tor were resistant to it. FK phage was entirely different from Mukerjee group IV phage in morphology and antigenicity. In addition to group IV phage, the use of FK phage will be useful in the examination and typing of *V. cholerae*.

Mukerjee cholera-typing phage of group IV was reported to lyse all strains of *Vibrio cholerae* biovar *cholerae*, but not to lyse strains of *V. cholerae* biovar El Tor (2). This was immediately confirmed by Takeya and Shimotori (3), and lysis by the group IV phage, in addition to the chicken erythrocyte agglutination and the polymyxin B sensitivity tests, has since been used as a valuable test for differentiating two biogroups of *V. cholerae* O:1 in most laboratories.

Recently, we succeeded in isolating a vibrio-infecting phage which was similar to the group IV phage in its lytic spectrum but different from it in morphological and serological properties.

Isolation. A new vibrio-infecting phage was

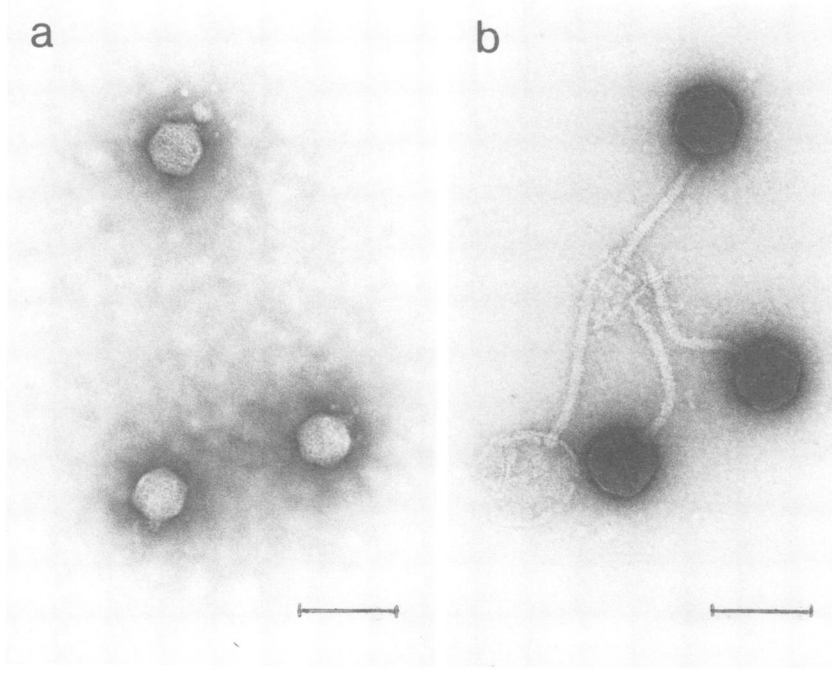

FIG. 1. *Electron micrograph of the FK phage (a) and the Mukerjee group IV phage (b). Bar, 100 nm.*

isolated from sewage by using the H218 SM^r strain of *V. cholerae* biovar *cholerae* as the propagating strain; it was named FK phage because it was isolated in Fukuoka City.

Lytic spectrum. All 25 strains of *V. cholerae* biovar *cholerae* were lysed by 10× routine test dilution of FK phage, whereas all 56 strains of *V. cholerae* biovar El Tor and 37 strains of *V. cholerae* non-O:1 were resistant to it. Strains of *V. cholerae* examined were collected from diverse sources, as shown in Table 1. Other species of bacteria, such as *Vibrio parahaemolyticus* (35 strains) and those of *Salmonella* (24 strains), *Shigella* (21 strains), *Escherichia* (8 strains), *Proteus* (12 strains), *Pseudomonas* (3 strains), *Bacillus* (4 strains), *Micrococcus* (1 strain), *Bordetella* (1 strain) and *Staphylococcus* (21 strains) were insensitive to FK phage, even at 100,000× routine test dilution.

TABLE 1. *Sources of cultures of V. cholerae*

Cultures	Place of isolation	Year(s) of isolation	No. of strains examined
V. cholerae biovar *cholerae*	Japan	1910s	3
		1962	1
	Bangkok	1947	3
		1954	2
		1958	1
	Culcutta	1956	4
	India	1957	1
		1960	4
	Bangladesh	1976	2
		1979	4
V. cholerae biovar El Tor	El Tor	1905?	2
	Celebes	1940?	2
		1959	1
		1961	2
	Culcutta	1958	1[a]
		1960	2
	Bangkok	1959	3[a]
		1960	1
		1978	6
	Ubon	1960	4
	Philippines	1961	3
		1978	5
	Japan	1962	3
		1963	1
		1976	2
	Guam	1974	10[b]
	Brazil	1978	4[a]
	Bangladesh	1978	2
		1979	2
V. cholerae non-O:1	Sudan	1968	12
	Bangladesh	1976	10
		1977	14
		1978	1

[a] Isolated from sewage water. All strains not footnoted in the table were isolated from human materials.
[b] Five strains were isolated from water sources.

Morphological properties. The FK phage is morphologically different from the group IV phage, as shown in the electron micrographs of Fig. 1. The head of the FK phage is 60 to 68 nm in diameter, and the tail is extremely small, 15 nm in length and 12 nm in width. On the other hand, the head of the group IV phage is 62 to 71 nm in diameter, and the tail is long, 160 to 170 nm in length and 13 to 15 nm in width.

Antigenic properties. The antigenic properties of FK and group IV phages were comparatively studied by serological neutralization tests with each rabbit phage antiserum. In cross-reactions, each phage was neutralized only by its homologous antiserum and never by its heterologous antisera. In other words, the two phages are antigenically different from each other.

Cross sensitivity test. Mutants resistant to FK, group IV, and kappa-type phages (3, 4) were selected, respectively, from the cells of their respective propagating strains to study the relation among these three phages. The results of cross sensitivity tests of each mutant to each phage revealed that no relation exists among the three phages (Table 2).

TABLE 2. *Cross sensitivity test of each phage-resistant mutant to each phage*

V. cholerae mutant strain	FK phage	Phage IV	Kappa-type phage
H218/FK	−	+	+
H218/IV	+	−	+
H218/K	+	+	−

The above-described data indicate that the FK phage, in addition to the group IV phage, can be routinely used in differentiating the two biotypes of *V. cholerae* O:1. In some experiments (S. Shimotori and K. Takeya, unpublished data), four English strains, which were isolated from an agricultural drainage ditch (1) and were resistant to polymyxin B but were lysed by group IV phage, were resistant to FK phage. Since there exist a few discrepancies in the lytic patterns of both phages, the use of FK phage in addition to group IV phage will be useful, especially in the examination of strains of *V. cholerae* with atypical biological properties.

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