A Transducing Bacteriophage for *Proteus rettgeri*

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Transducing phages are usually derived directly from lysogenic strains (Zinder & Lederberg, 1952; Lennox, 1955; Morse, Lederberg & Lederberg, 1956; Coetzee, de Klerk & Smit, 1967), but have been isolated from soil (Thorne, 1962; Takahashi, 1963) and sewage (Holloway & van de Putte, 1968). The incidence of lysogeny in *Proteus rettgeri* is reported as 0% (Tauebeneck, 1962), 22.7% (Coetzee, 1963) and 2.7% (Vieu, 1963). Phage active on *P. rettgeri* is readily isolated from sewage (Coetzee, 1963), and it was decided to try this source, as well as lysogenic strains, in a search for transducing phages for *P. rettgeri*.

The methods have been described (Coetzee & Sacks, 1960; Coetzee, 1963; Coetzee, Smit & Prozesky, 1966). One hundred and fifty recently isolated strains of *Proteus rettgeri* were treated with mitomycin C (Kyowa Hakko Kogyo Co. Ltd, Tokyo) (Seaman, Tarmy & Marmur, 1964). The culture supernatants were sterilized with chloroform and spotted on all the strains. Twenty-seven strains (18%) proved lysogenic. Phage isolated from sewage lysed 98 of 144 of the strains employed. Clear-plaque-forming phage may transduce (Takahashi, 1963), but screening for transducing phages was restricted to those which produced turbid plaques (Boyd, 1951). Eleven sewage phages and 8 phages from lysogenic *P. rettgeri* were examined. Only the phage (7/R49), derived from *P. rettgeri* strain R7, transduced markers into its indicator organism R49. It transduced prototrophy to R49 *try-1* at a rate of 4 × 10^{-7} p.f.u. adsorbed. Adsorption was not Ca^{2+} dependent, and the clear plaques developed turbid centres after 48 hr.

Phage 7/R49 differs morphologically (Pl. 1) from other *Proteus* transducing phages (Prozesky, de Klerk & Coetzee, 1965; Coetzee *et al.* 1966; Coetzee *et al.* 1967). It has an hexagonal head, a neck and an unsheathed tail, which ends in a number of fibres. The over-all length is 2310 Å. It resembles a number of other *P. rettgeri* phages (see Fig. 22, 27, 28, Prozesky *et al.* 1965).

Strain R49 is a typical *Proteus rettgeri* (Rauss, 1962). Bacterial DNA was extracted (Johnson & Ordal, 1968) and the molar content of guanine + cytosine (GC%), calculated from its thermal denaturation temperature (Marmur & Doty, 1962), was 44%. Hill (1966) has reported the GC% of one strain of *P. rettgeri* as 39.5 to 42%, depending on the method employed. The GC% of the phage 7/R49 DNA, extracted by the method of Davison & Freifelder (1962) and determined as above, was 47%. This disparity may indicate that the bacterial DNA was incorporated into transducing particles of phage 7/R49 without integration in the viral DNA (Okubo *et al.* 1963). The GC% of DNA of other *Proteus* transducing phages and their bacterial hosts correspond closely (Coetzee *et al.* 1966; Coetzee *et al.* 1967).

It was found by Arber (1960) with coliphage P1, that small doses of ultraviolet irradiation applied to phage lysates caused an increase in transductants, followed by an exponential decrease with larger doses. The plaque-forming titre of the phage declined progressively at a greater rate. This differential effect of ultraviolet light on plaque formation and transduction was encountered in the transducing phages of *Proteus*
Phage 7/R49 suspended in 0.1 M ammonium acetate (pH 7.2) and negatively stained with 1.0% (w/v) potassium phophotungstic acid.
mirabilis and Providence (Coetzee & Sacks, 1960; Coetzee et al. 1966), but not in those of P. morganii (Coetzee et al. 1966) or P. vulgaris (Coetzee et al. 1967). All transductants were lysogenic for phage 7/R49. The phage could transduce the markers try-1, leu-1; try-1, pro-1; try-1, arg-1 in successive steps to multiple auxotrophs of R49. Primary selection was on minimal medium supplemented with one of the amino acids and resulting clones were again exposed to the transducing phage with selection on minimal medium. The rate of the later transduction was about $3 \times 10^{-1}$ of the earlier (Coetzee et al. 1967).

Many Proteus rettgeri phages plate on P. hauseri and Providence strains (Coetzee, 1963), but phage 7/R49 was not active on any of 35 Providence, 30 P. morganii, 45 P. mirabilis and 160 P. vulgaris strains. Phages are now available which transduce markers within each species of the Proteus-Providence group, but, owing to the restricted host range of these phages, only limited interstrain (Coetzee & Sacks, 1960) and no inter-species hybridization is possible. These phages are being used for genetic mapping of P. mirabilis (Prozesky, 1969) and other Proteus species. It is hoped, ultimately, to compare the chromosomes of the different species.

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REFERENCES


Short communications


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