Abortive Transduction of Motility in *Proteus* and *Providencia* Strains

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SUMMARY

Abortive transduction of motility has been demonstrated with all members of the *Proteus-Providencia* group of bacteria.

INTRODUCTION

Coetzee (1963) transduced the swarming property into non-swarming mutants of *Proteus mirabilis* with phage 34. No abortive transductants in the form of trails (Lederberg, 1956; Stocker, 1956) were observed. Similarly, no abortive transduction (Ozeki, 1956) was encountered in auxotroph to prototroph transductions with *P. mirabilis* 13 *arg H2*, *metE-1*, *cyseE-6*, *trp-1* using phage 34.13 (Prozesky, 1968; Grabow, 1970); *Proteus vulgaris* 69 *met-4* with phage 107/69 (Coetzee, De Klerk & Smit, 1967); *Proteus rettgeri* R49 *trp-1*, *leu-1*, *pro-1*, *arg-1* with phage 7/R49 (Krizsanovich, De Klerk & Smit, 1969); *Proteus morganii* 2815 *hi-1*, *ad-1*, *trp-1* with phage M (Coetzee, Smit & Prozesky, 1966); and *Providencia* 9295 *ad-4*, *cys-10*, *leu-1* with phage PL25 (Coetzee *et al.* 1966).

Böhme (1964) suggested the occurrence of abortive transduction to explain results obtained in transducing methionine auxotrophs of *Proteus mirabilis*, although no microcolonies were seen.

It was felt that if abortive transduction can occur in these strains the best way to demonstrate it would be by transduction of motility.

METHODS

Bacteria and phages. These are listed in Table 1. The *fla* mutants were selected and independently isolated on semi-solid medium (0.35 % agar) after treatment of swarming strains with *N*-methyl-*N'*-nitro-*N*-nitrosoguanidine (Adelberg, Mandel & Chen, 1965). Non-swarming colonies were picked off on to the same medium and further investigated. Absence of flagella was confirmed by their failure to agglutinate with homologous H antisera and by electron microscopy. Phage lysates were prepared according to the method of Coetzee & Sacks (1960a).

Media. Difco nutrient broth was used and the semi-solid medium contained 3.5 g Difco agar, 10 g Bacto Tryptone and 6 g NaCl/l distilled water. Plates were dried with their lids removed for 1 h at 37°.

Fig. 1, 2. Preparations were obtained by incubating 0.5 ml of overnight broth culture with 0.5 ml phage (5×10^9 p.f.u./ml). Adsorption was for 15 min at 30° . Mixtures were streaked on to semi-solid agar plates and incubated overnight. Arrows indicate swarms (S), trails (T) and branched trails (B).

Fig. 1. *Proteus mirabilis* *fla*-7 transduced to motility with wild-type ϕ 34. Numerous trails and some swarms are seen.

Fig. 2. *Proteus rettgeri* *fla*-1 transduced to motility with wild-type ϕ 7/R49. A few swarms and trails (some of which are branched) are seen.

Transduction techniques. These were as described by Coetzee (1963) except for the composition of the agar (0.35 %) and the incubation temperature, which was 30° . Plates were incubated for 18 to 24 h.

RESULTS AND DISCUSSION

In transduction of *fla* recipients with wild-type phage, fan-shaped swarms and linear arrays of colonies which extended a few millimeters away from the inoculum were invariably formed (Fig. 1, 2). These trails reached maximum length after about 18 h incubation. Occasionally a swarm arose from one (often the most distal) of the colonies in a trail.

Trails were usually least frequent in the *Proteus rettgeri*, *P. vulgaris* and *Providencia* strains and most abundant in *P. mirabilis* and *P. morganii*, where they often far outnumbered the

Table 1. *Proteus* and *Providencia* transducing systems

Parent strain	Mutants	Phage	Reference
<i>Proteus mirabilis</i> 13	<i>fla</i> 1-13	34	Coetzee & Sacks (1960a)
<i>Proteus morganii</i> 2815	<i>fla</i> 1-5	M	Coetzee <i>et al.</i> (1966)
<i>Proteus rettgeri</i> R49	<i>fla</i> 1, 2	7/R49	Krizsanovich <i>et al.</i> (1969)
<i>Proteus vulgaris</i> 69	<i>fla</i> 1, 2	107/69	Coetzee <i>et al.</i> (1967)
Providencia 9295	<i>fla</i> 1-5	PL 25	Coetzee <i>et al.</i> (1966)

swarms. Colonies picked from trails and inoculated separately on agar either developed into a single colony or swarmed. The trails resembled the abortive transduction of motility described by Stocker (1956) and Lederberg (1956) in *Salmonella* and by Armstrong & Adler (1967) in *Escherichia coli*. Some trails branched, for which a possible explanation is that daughter cells without the non-replicating transduced chromosome fragment inherited sufficient wild-type gene product to render them motile in the very soft agar (Stocker, 1956). In transduction between mutants in the *P. mirabilis*, *P. morganii* and *Providencia* strains some crosses yielded swarms but no trails.

In such cases the lesions in the donor and recipient genomes could have been in the same cistron. This effect was usually reciprocal, but a few mutants of *Proteus mirabilis* showed non-reciprocal complementation. This phenomenon has also been encountered by Armstrong & Adler (1967) in *Escherichia coli* and by Iino & Enomoto (1966) in *Salmonella*. Using the mutants listed in Table 1 it was possible provisionally to divide the *fla* mutants of both *P. mirabilis* and *Providencia* into four complementation groups while the *fla* mutants of *P. morganii*, *P. vulgaris* and *P. rettgeri* each fell into two groups.

Two types of controls were employed. In the one the phage used was prepared on the recipient and in the other the phage-suspension was replaced by an equal volume of broth. These usually produced no swarms. Occasionally broth control plates of *Proteus mirabilis* and *P. vulgaris fla* mutants showed a few trails. This occurred in the absence of phage – a situation similar to that encountered by Armstrong & Adler (1967) with some *mot* mutants of *Escherichia coli*.

Quadling & Stocker (1957) demonstrated that certain *Proteus* 'O' strains gave rise to satellite micro-colonies, and the existence of the 'Tröpfchenform' colonies of *Proteus hauseri* (Coetzee & Sacks, 1960b) may be a similar phenomenon. The occurrence of these spontaneous trails does not reduce the significance of trails encountered in the transduction experiments, which were always much more numerous and reproducible. Thus in experiments with *P. mirabilis* or *P. vulgaris* it was possible to divide *fla* mutants into groups which consistently gave 0 to 50, 50 to 250 or 250 to 500 trails with phage prepared on other mutants.

Failure to detect trails in the previous *Proteus mirabilis* experiments (Coetzee, 1963) may have been due to the higher agar concentration (0.6 %) and the higher incubation temperature (37°) used. Both of these variables are known to affect flagellar formation and function in *Proteus* spp. (Coetzee & De Klerk, 1964).

We have now demonstrated the abortive transduction of motility with all members of the *Proteus*-*Providencia* group, but the reason for failure to demonstrate the phenomenon with other markers has to be determined.

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