Arginine Gene Clusters in the *Proteus*-*Providencia* Group

By O. W. PROZESKY, W. O. K. GRABOW,* SUSANNA VAN DER
MERWE AND J. N. COETZEE

Department of Microbiology, University of Pretoria,
Pretoria, South Africa

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Five of eight structural genes for arginine synthesis in *Proteus mirabilis* strain 13 are closely linked in the order *argECBGH* (Prozesky, 1968*a*). These genes code for the fifth, third, second, seventh and eighth enzymes of the pathway respectively (Prozesky, 1967). *Escherichia coli* and *Salmonella typhimurium* have an analogous cluster which consists of four genes *argECBH* (Glansdorff, 1965; Sanderson, 1970). The arginine genes of *Pseudomonas aeruginosa* are not linked by transduction (Feary, Williams, Calhoun, & Walker, 1969). Gene topography and clustering are related to gene regulation and evolution (Bodmer & Parsons, 1962; Hedges, 1972) and regulation of the cluster differs in *Proteus mirabilis* and *E. coli* (Cunin *et al.* 1969; Prozesky, 1969). These five genes and where possible the linked external marker *cysE* (Böhme, 1964; Grabow, 1972) were mapped in strains of *Proteus* and *Providencia* for which transducing phages are available (Coetzee, 1972). Knowledge of the relative topography is important for taxonomy and studies on gene regulation and evolution (Prozesky, 1969; Baumberg & Ashcroft, 1971; Krzyzek & Rogers, 1972; Hedges, 1972).

Media and supplements were as described (Prozesky, 1967; 1968*a, b*) except that L-methionine (0.0001 %) replaced L-cysteine in minimal medium for *Proteus morganii* (Coetzee, Smit & Prozesky, 1966). The bacterial strains and the transducing phages have been described by Coetzee (1972). The mutants (Table 1) were those of Prozesky (1967, 1968*a*) or mutants isolated by treatment with *N*-methyl-*N'*-nitro-*N*-nitrosoguanidine (Adelberg, Mandel & Chen, 1965; Childs & Smith, 1969). Biochemical blocks were determined as before (Prozesky, 1967; Grabow, 1972) in cell-free extracts and urease negative mutants of *Proteus mirabilis* were isolated and characterized as described by MacLaren (1968). The newly isolated arginine mutants conform to those of Prozesky (1967) except that the *Proteus rettgeri* and *P. morganii* mutants blocked prior to *argH* are permeable to and respond to L-arginino-succinate (Prozesky, 1967). Transductions were performed at a phage input multiplicity of about 5 (Prozesky, 1968*a*). Mutations were mapped by ratio tests with linked markers within the cluster (Glansdorff, 1965; Prozesky, 1968*a*) and with the linked external marker *cysE* (Prozesky, 1968*a*). The latter mutation could not be mapped in *P. morganii* as the wild-type is cysteine or methionine dependent (Porter & Meyers, 1945). No *cysE* mutants could be isolated from the *P. rettgeri*. A minimum of 1000 transductants were scored for each cross in duplicate experiments. Intermutation distances were calculated according to Prozesky (1968*a*) and mean values of reciprocal crosses were taken as the distance between mutations (Prozesky, 1968*a, b*).

Results are presented in Table 1. *Proteus mirabilis*, *P. vulgaris*, *P. rettgeri* and *P. morganii* have the five-gene *argECBGH* cluster peculiar to *Proteus* (Prozesky, 1968*a*). In the first two

* Present address: National Institute for Water Research, South African Council for Scientific and Industrial Research, Pretoria.

Table 1. *Bacteria, phages and results of transduction experiments*

Organism	Mutants*	Trans- ducing phage	Urease†	Cluster	Minimum cluster size	Minimum distance, <i>argE-cysE</i>	Reference
<i>Proteus mirabilis</i> 13	<i>argB1</i> , <i>C1</i> , <i>E1</i> , <i>G1</i> , <i>H1</i> , <i>cysE1</i> , <i>ure1</i> , <i>ure2</i> .	34	+	<i>argECBGH</i>	19.3‡	76.8‡	Prozesky (1968 <i>a</i>)
<i>P. rettgeri</i> R49	<i>argB1</i> , <i>B2</i> , <i>C1</i> , <i>C4</i> , <i>E1</i> , <i>E2</i> , <i>G1</i> , <i>G2</i> , <i>H1</i> , <i>H2</i>	7-R49	+	<i>argECBGH</i>	22.0	No <i>cysE</i> isolated	van der Merwe (1972)
<i>P. vulgaris</i> 69	<i>argB1</i> , <i>B2</i> , <i>C1</i> , <i>C2</i> , <i>E1</i> , <i>E2</i> , <i>G1</i> , <i>G2</i> , <i>H1</i> , <i>H2</i> , <i>cysE1</i>	107-69	+	<i>argECBGH</i>	21.9	78.0	Coetzee (1972)
<i>P. morganii</i> 2815	<i>argB1</i> , <i>C1</i> , <i>C2</i> , <i>E1</i> , <i>E2</i> , <i>G1</i> , <i>G2</i> , <i>H1</i> , <i>H2</i>	M	+	<i>argECBGH</i>	22.5	Wild-type cysteine- dependent	Coetzee (1972)
<i>Providencia</i> 9295	<i>argB1</i> , <i>B2</i> , <i>C1</i> , <i>C2</i> , <i>E1</i> , <i>E2</i> , <i>G1</i> , <i>G2</i> , <i>H1</i> , <i>H3</i> , <i>cysE1</i> , <i>cysE2</i>	PL25	-	<i>argECBH</i>	17.3	Not linked	Coetzee (1972)

* Enzyme deficiencies: *argB*, ATP: α -N-acetylglutamate-5-phosphotransferase; *argC*, α -N-acetyl-L-glutamate- γ -semialdehyde: NADP oxidoreductase, EC. 1.2.1.4; *argE*, α -N-acetyl-L-ornithineamidohydrolase, EC. 3.5.1.1; *d*, *argG*, L-citrulline:L-aspartate-lyase, EC. 6.3.4.5; *argH*, L-argininosuccinate-L-arginine lyase, EC. 4.3.2.1; *cysE*, O-acetylserine sulphydrylase; *ure*, urea amidohydrolase, EC. 3.5.1.5.

† Presence or absence of urease in wild-type.

‡ Calculated as $\frac{\text{wild-type transductants}}{\text{total transductants}} \%$.

organisms the cluster is linked by transduction to *cysE* (Table 1; Prozesky, 1968*a*). Providence has a four-gene *argECBH* cluster not linked to *cysE*. In all enterobacteria examined possession of urease (Coetzee, 1972) accompanies an *argECBGH* cluster. Absence of this enzyme is connected with the *argECBH* cluster. The *ure* gene is not linked to the gene group in *P. mirabilis*, the only organism investigated for this property.

An *argECBGH* topography in bacteria equipped with a potent arginase-urease degradation pathway may serve an unknown regulatory function not present in urease negative organisms (Prozesky, 1968*b*). Arginine itself is not a potent end-product repressor of the biosynthetic pathway in *Proteus mirabilis* and the true metabolic regulation of arginine synthesis remains unknown (Prozesky, 1968*b*, 1969). The finding that the *ure* gene for urease is not linked to the cluster nor to any other arginine genes in *P. mirabilis* (Prozesky & Coetzee, unpublished) means that *ure* is not included in an arginine 'operon'. The order of the *argECBH* genes in both types of cluster is the same even in widely divergent bacteria. The significance of this order is unknown but may be a requisite for regulation (see Baumberg & Ashcroft, 1971). It is interesting that *Proteus morganii* with a guanine plus cytosine molar content (G+C%) equivalent to that of *Escherichia coli* and *Salmonella* (Hill, 1966) has the *argECBGH* 'Proteus' arrangement and a urease. In contrast Providence has a G+C% similar to that of the remaining members of the *Proteus* group but lacks a urease and has the *argECBH* cluster. These findings as well as the biochemical and serological differences among these species (Coetzee, 1972) indicate that the respective clusters have been retained in spite of development of other relatively large genetic differences. We agree with Hedges (1972) that the different gene clusters may represent stages in the evolution of an operon, and postulate that selection pressure for this cluster may be connected with methods of arginine and urea catabolism (Prozesky, 1968*b*).

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