Expression of Escherichia coli K-12 Arginine Genes in Pseudomonas fluorescens

M. MERGEAY,1* A. BOYEN,2 C. LEGRAIN,3 AND N. GLANSDORFF2

Département de Radiobiologie, Centre d’Etudes de l’Energie Nucléaire, C.E.N./S.C.K., Mol1; Laboratorium voor Erfelijkheidsleer, Vrije Universiteit te Brussel2; and Laboratoire de Microbiologie, Université Libre de Bruxelles, Bruxelles, Belgium3

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Various F' plasmids have been transferred from Escherichia coli K-12 into Pseudomonas fluorescens strain 6.2 (7). In particular, the F110 episome carrying the argECBH divergent operon (4, 5) was found to complement the arg-122 and met-13 mutations of P. fluorescens strain PMG122; the latter mutations were therefore assumed to be argH and metaA, respectively. We have investigated the expression and the control of E. coli arg genes in a PMG122 (F110) transconjugant, taking advantage of the fact that ornithine biosynthesis proceeds differently in Enterobacteriaceae and Pseudomonadaceae (9).

In E. coli, ornithine and acetate are the products of the deacetylation of N-α-acetylornithine catalyzed by N-α-acetylornithinase (EC 3.5.1.16) (Fig. 1, pathway A). In Pseudomonas, ornithine is produced by an energy saving cycle: the acetyl group of N-α-acetylornithine is transferred onto glutamate by glutamate-ornithine acetyltransferase (EC 2.3.1.35) giving N-α-acetylglutamate, a precursor of acetylornithine (Fig. 1, pathway B).

Assays of N-α-acetylornithinase (argE gene product), argininosuccinate lyase (EC 4.3.2.1., coded for by argH), and the acetyltransferase have been performed as reported previously (6) on cultures grown at 29°C in minimal medium (7) supplemented or not with arginine. For the transconjugant, omitting the methionine ensured a selective pressure maintaining the plasmid. Results are compared in Table 1 with specific activities found in E. coli grown in various repression conditions. The following conclusions can be drawn. (i) The glutamate-ornithine acetyltransferase is present in P. fluorescens. (ii) As expected from genetic tests, the argininosuccinate lyase is defective in PMG122. A functional enzyme is present in the transconjugant. (iii) E. coli acetylornithinase is made in the transconjugant, in addition to the host acetyltransferase. A weak “acetylornithinase” activity is observed in PMG122; this may be due to a carboxypeptidase, as is the case in Saccharomyces cerevisiae (2) and Thermus aquaticus (3).

The results amply confirm the conclusion, previously based on genetic data only, that metabolic deficiencies of P. fluorescens can be corrected by E. coli genes brought in on a F-plasmid vector (7). Synthesis of N-α-acetylornithinase, an enzyme alien to Pseudomonas, demonstrates this unambiguously.

The E. coli genes are not repressible by arginine in P. fluorescens, as expected from fact that the only known trans-acting regulatory gene of the arginine regulon is not carried by plasmid F110. E. coli trp genes disconnected from their control region and transferred into P. aeruginosa on a RP4 vector (8) are expressed in their new host and are, of course, not repressible.

The specific activities of the E. coli argE and

![Fig. 1. Biosynthetic pathways for ornithine in E. coli (A) and in Pseudomonas (B).](http://jb.asm.org/ on September 28, 2017 by guest)
argH enzymes in Pseudomonas are close to the values found for wild-type E. coli cells synthesizing arginine endogenously; this amounts to a third (for argE) or a fifth (for argH) of the potential of genetically derepressed cells (Table 1; compare line 2 with line 4). It appears, therefore, that no drastic restriction limits the expression of E. coli genes in P. fluorescens or the functioning of the corresponding proteins. Nor does the mechanism of divergent transcription of argECBH appear to require an element coded for by a gene extraneous to the chromosomal region carried by F110. The successful complementation of E. coli argH mutants by cloned yeast DNA (1) provides another case of expression of an argH gene in a foreign cytoplasm.

LITERATURE CITED