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NOTES

Location of the Structural Gene for Fructose-1,6-Diphosphate Aldolase in *Escherichia coli*

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Gene *fda* has been mapped, by co-transduction, between *thyA* and *serA* on the *Escherichia coli* chromosome.

In previous experiments, the structural gene for fructose-1,6-diphosphate aldolase (*fda*) has been mapped by non-interrupted mating procedures and has been located approximately in the *str-serA* region of the *Escherichia coli* chromosome (1). Recently, Portalier et al. (4) reported a similar map position of *fda*, but they were unable to demonstrate co-transduction of *fda* with several of the known markers in this chromosome region although the markers used by them form a chromosomal segment continuously linked by P1 transduction. We have therefore attempted the precise mapping of *fda* to determine any relation of its position with the location of other enzymes of intermediary metabolism (5).

At first, preliminary mapping experiments were performed and the following results were obtained. (i) Neither of the Hfr strains, KL 16 or AB 2297, transferred *fda* as an early marker, thus restricting its location to the chromosomal segment between the points of entry of the chromosomes of these two strains, namely, to the region between min 55 and min 60. (ii) Interrupted mating experiments between Hfr J4 and a derivative of strain NP 315 (*fda*$^-$ *metC*$^-$

TABLE 1. *Localization of the fda marker by co-transduction*[a]

Donor		Recipient		Selected marker	No. of recombinants analyzed	Nonselected marker	Frequency of nonselected marker
Strain	Genotype	Strain	Genotype				
MA 197	*thr leu serA speB thi argR str*^{R} F$^-$	NP 315	*fda str*^{S}	*fda*$^+$	74	*serA*$^+$ *speB*$^+$ *serA*$^-$ *speB*$^+$ *serA*$^+$ *speB*$^-$ *serA*$^-$ *speB*$^-$	31 38 2 3
NP 315	*fda str*^{S}	AB 2295	*leu metB argG his thyA str*^{R}	*thyA*$^+$	195	*thyA*$^+$ *fda*$^-$ *thyA*$^+$ *fda*$^+$	124 71
NP 315	*fda str*^{S}	DR 24	*thr leu serA nalA*^{R} *str*^{R}	*serA*$^+$	52	*serA*$^+$ *fda*$^+$ *serA*$^+$ *fda*$^-$	25 27

[a] Transduction experiments were performed according to Lennox (2). Lysates of phage P1 were obtained by at least two passages of the phage in the respective donor strain. Recombinants were routinely checked for maintainance of the nonselected and unlinked genes. Ten transductants each of the genotype *serA*$^+$ *fda*$^+$ and *serA*$^+$ *fda*$^-$ of the third cross of Table 1 were checked for fructose-1,6-diphosphate aldolase activity in crude cell-free extracts; wild-type like aldolase activity and temperature-resistant growth were fully correlated. Strain DR 24 was made by mating Hfr KL 16 *nalA*^{R} with strain MA 197(3) and selecting *nalA*^{R} *str*^{R} recombinants.

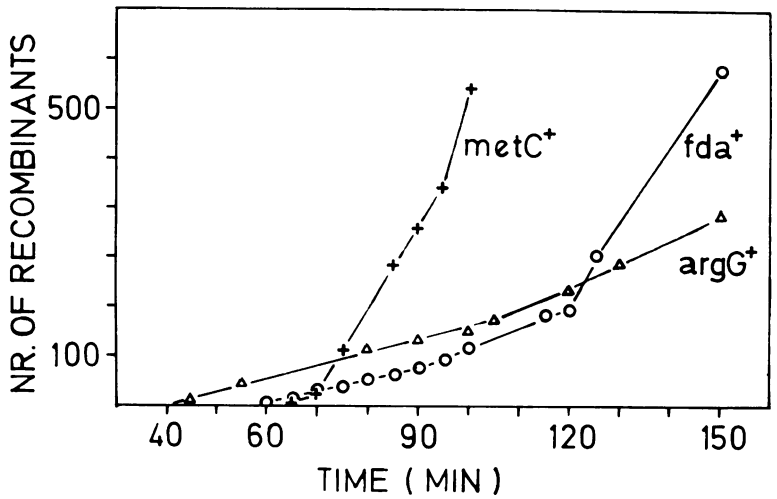

FIG. 1. *Interrupted mating experiment between Hfr J4 and strain NP 315 fda⁻ argG⁻ metC⁻ str^R (this strain was constructed by mating strain NP 315 first with strain AB 2295 and subsequently mating one of the resulting fda⁻ argG⁻ recombinants with Hfr strain AT 2446 thi-1 metC69 rel-1 to further introduce the metC marker). The fda⁺ genotype was selected by incubating plates containing glucose as carbon source at 40 C after allowing a phenotypic lag of 6 h at 30 C.*

argG⁻ str^R) indicated a map location of *fda* close to *argG* and *metC* (Fig. 1); however, in several experiments no clear-cut entry times of *fda* could be obtained.

The mapping of the *fda* locus was, therefore, approached by testing for co-transduction of *fda* with any of the known markers of the map segment in question. In accordance with the results of Portalier et al. (4), no joint transduction could be established with the following markers: *argG*, *uxaA*, *metC* and *tolC*. On the other hand, co-transduction with *serA* was observed (Table 1). If strain MA 197(3) was used as donor to transduce temperature-resistant growth into strain NP315 *fda⁻* the markers *serA* and *speB* were co-transduced with a linkage of 55 and 6%, respectively. The frequencies of the recombinant classes obtained (Table 1) indicate a location of *fda* on the counter-clockwise side of *serA* as the map is usually presented. A co-transduction of *fda* with *thyA* was therefore feasible and could indeed be demonstrated (frequency 64%) in a transduction experiment with strain NP 315 as donor and strain AB 2295 as recipient. As a control a reciprocal cross was performed. Serine prototrophy was introduced into strain DR 24 from strain NP 315, and a similar coinheritance of *fda* and *serA* (52%) could be observed. (Table 1) By using the formula of Wu (6) for relating transduction frequencies with map distances in minutes, the location of the *fda* gene was calculated (Fig. 2).

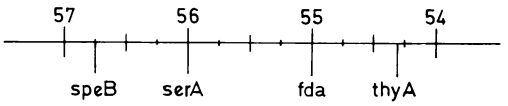

FIG. 2. *The section of the E. coli chromosome from min 54 to min 57, showing the relation of the fda map position to the markers used in this study.*

No linkage to any other structural gene of enzymes of the intermediary metabolism mapped until now is observed.

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LITERATURE CITED

1. Böck, A., and F. C. Neidhardt. 1966. Isolation of a mutant of *Escherichia coli* with a temperature-sensitive fructose-1,6-diphosphate aldolase activity. J. Bacteriol. **92**:464-469.
2. Lennox, E. S. 1955. Transduction of linked genetic characters of the host by bacteriophage P1. Virology **1**:190-206.
3. Maas, W. K. 1972. Mapping of genes involved in the synthesis of spermidine in Escherichia coli. Mol. Gen. Genet. **119**:1-9.
4. Portalier, R. C., J. M. Robert-Baudouy, and F. R. Stoeber. 1972. Localisation génétique et caractérisation biochimique de mutations affectant le gène de structure de l'hydrolase altronique chez Escherichia coli K 12. Mol. Gen. Genet. **118**:335-350.
5. Taylor, A. L., and C. D. Trotter. 1972. Linkage map of *Escherichia coli* strain K-12. Bacteriol. Rev. **36**:504-524.
6. Wu, T. T. 1966. A model for three-point analysis of random general transduction. Genetics **54**:405-410.