PREFERENTIAL SYNTHESIS OF β-GALACTOSIDASE AFTER AMINO ACID STARVATION

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In a given medium, constituents of a bacterial cell increase at unique rates; sometimes this balance is broken under abnormal conditions. In the system reported here, β-galactosidase is synthesized at very high rate.

E. coli K-12 58-161, methionineless and inducible for β-galactosidase, synthesizes RNA even in the absence of an exogenous supply of amino acids (Borek et al, 1955). This character is controlled by a single gene called RC (Stent and Brenner, 1961). In a constitutive mutant of this strain, a very high initial rate of β-galactosidase synthesis was observed after methionine starvation in the presence and the absence of a carbon source (Fig. 1, Curve 5). In the original inducible strain, the synthesis of β-galactosidase is strongly repressed after methionine starvation if a carbon source is present during starvation; if the cells are starved for both methionine and a carbon source, the enzyme is synthesized even during starvation (Fig. 1, Curve 6, Yanagisawa 1962). In the inducible strain, therefore, a negative preferential synthesis was observed when methionine was restored. In the constitutive strain, total protein synthesis, studied by the uptake of C^{14}-leucine, did not show such an abnormal behavior (Fig. 2). This preferential synthesis of β-galactosidase is observed even after 10 minutes of methionine starvation.

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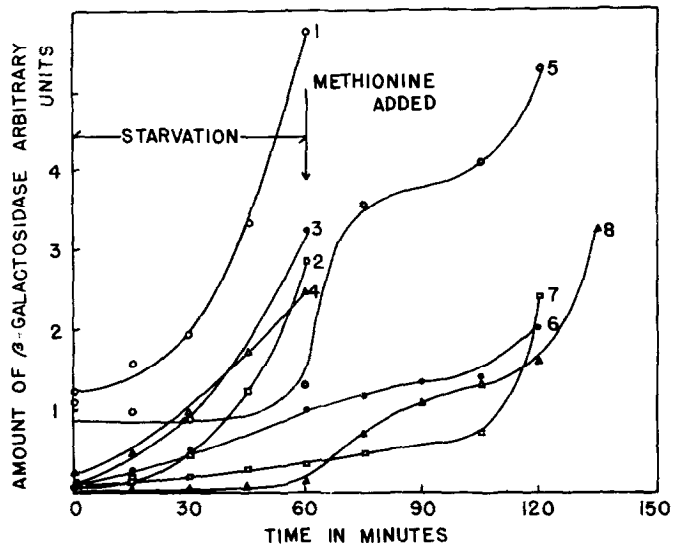

Fig. 1, Preferential synthesis of β-galactosidase after methionine starvation.
Curves 1-4. control: glycerol and methionine were present from O time. The inducer IPTG was given to the inducible cells at O time.

1. E. coli (K-12) 58-161, constitutive.
2. E. coli (K-12) 58-161, inducible.
3. E. coli 15, his⁻ met⁻, inducible.
4. E. coli (K-12) Ya-2, inducible.

Curves 5-8. Starved for methionine and glycerol in the presence of IPTG.

5. E. coli (K-12) 58-161, constitutive.
6. E. coli (K-12) 58-161, inducible.
7. E. coli 15, his⁻ met⁻.
8. E. coli (K-12) Ya-2.

As mentioned before, this strain synthesizes about 80% of the normal amount of RNA after 60 minutes starvation. As a control, E. coli 15, histidineless, methionineless and inducible for β-galactosidase, in which RNA synthesis is stringently controlled by amino acids, was used. As shown in Figure 1, curve 7, there was no preferential synthesis of β-galactosidase upon restoration of methionine. From these results, it is not clear whether the difference is caused by a different R.C locus in the two strains or by a difference in inducibility. The following facts seem to favor the former view.

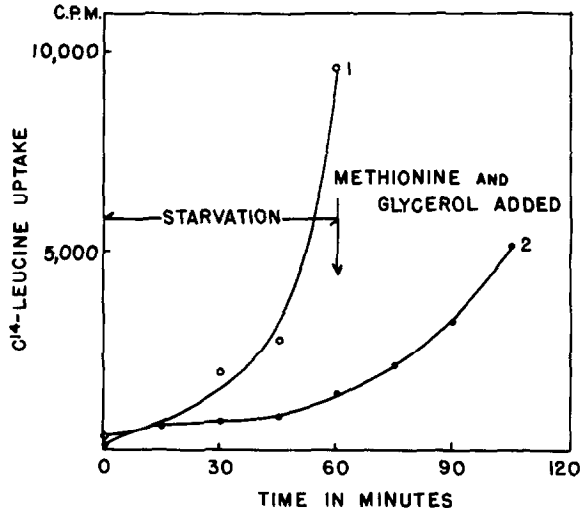

Fig. 2, C^{14}- leucine uptake by E. coli (K-12) 58-161, constitutive for β -galactosidase.

1. Control: glycerol and methionine were present from O time.
2. Starved for methionine and glycerol for 60 minutes.

E. coli K-12 Ya-2, methionineless and inducible for β-galactosidase, which was obtained by Stent and Brenner (1961) upon crossing 58-161 when the cells were deprived of both methionine and glycerol (Fig. 1, Curve 8). After 60 minutes of methionine starvation, β -galactosidase was synthesized at a high initial rate regardless of the presence or absence of inducer during starvation. However, the rate is much smaller in comparison with E. coli 58-161. In Ya-2, the synthesis of RNA is more stringently controlled by amino acids than in 58-161. The former synthesizes about 40% of the normal amount of RNA after 60 minutes of methionine starvation instead of the 80% observed in 58-161. Therefore, it would seem that this preferential synthesis of the enzyme has some relation to the state of RC locus but not a direct relation to the accumulation of RNA during starvation, since in the absence of a carbon source RNA did not accumulate.

Acknowledgement

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