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Formation of non-phage-antigenic protein in *E. coli* infected with T2 phage*

It has been found that the integrity of the infecting phage deoxypentose nucleic acid (DNA) is required for the successful initiation of the syntheses of both phage-specific protein (phage antigen)¹ and DNA. Recently, a possibility that the parental phage DNA controls the formations of phage antigen and DNA *indirectly* through a substance other than DNA has been suggested^{1, 2}. If the control by the parental DNA is really indirect, there must be a formation of a new substance or substances immediately after the infection but before the appearances of phage DNA and antigenic protein. The present note describes some of our experimental results on a protein formed just after infection which are in favour of the hypothesis of indirect control by the parental DNA.

The formation of protein in *Escherichia coli*, strain B(H), infected with bacteriophage T2r⁺ was studied in the lysates at different times of phage development. T2-antigenic protein is defined here as that protein which can be precipitated specifically with anti-T2 rabbit serum¹. The result of a typical experiment is presented in Fig. 1. It is clear that there is a very active protein synthesis immediately after infection, confirming the results of previous workers³, and that the protein formed before 10 min after infection does not contain T2-antigenic protein. The rate of the formation of this non-phage-antigenic protein is high for the first 10 min and then gradually tapers off.

It was found that the non-phage-antigenic protein is not a precursor protein of phage antigen, because most of the ³⁵S (more than 90 %) in non-phage-antigenic protein fraction, which had been labeled by feeding ³⁵S only during the first 8 min after infection, could not move into the phage-antigenic protein fraction even after 60 min incubation in ³⁵S-free medium.

The next question is whether it is a normal bacterial protein or not. The following evidences suggest that it is not bacterial. (1) The fact that the infection with T2 phage can provoke a synthesis of the non-antigenic protein in ultraviolet light (UV) irradiated *E. coli*, in which a synthesis of bacterial protein is largely suppressed (see Fig. 2). (2) A similar observation that the adenine-requiring mutant of *E. coli* can form the non-antigenic protein immediately after infection even in a medium without adenine, while it can hardly synthesize protein in this medium if it is not infected (I. WATANABE AND Y. KIHO, unpublished). From these results, it can be considered that the non-antigenic protein is different from the bacterial protein which can be produced in uninfected normal coli. (3) The fact that protein synthesis is prerequisite to the formation of new phage DNA after infection⁴. This fact indicates the presence of a new kind of protein in the

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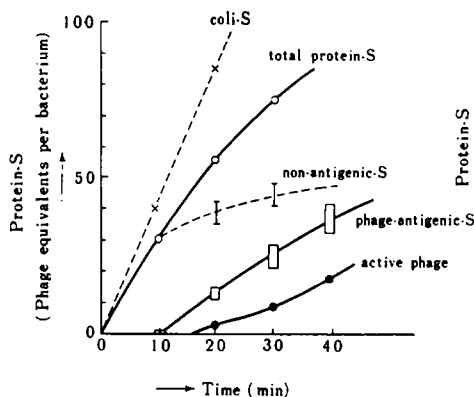


Fig. 1. Protein synthesis in *E. coli* after infection with T₂ phage. ³⁵S was added in tris-glucose medium¹ at zero time after infection with T₂ and the measurements of ³⁵S activities were made on the cyanide lysates at different times. The crude radioactive lysate was first dialyzed against adsorption buffer and then centrifuged at low speed. The supernatant was assayed for infective phage, total and T₂-antigenic proteins¹. In this experiment, total ³⁵S activity in this supernatant was conveniently regarded as total protein ³⁵S. The amounts of proteins are expressed in multiples of the amount of sulfur per T₂ phage particle ($2.4 \cdot 10^{-12} \mu\text{g}^1$).

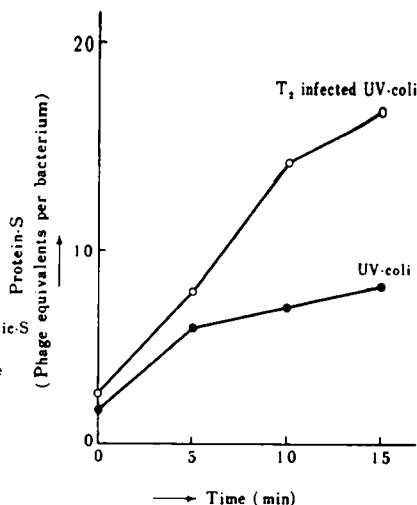


Fig. 2. Protein synthesis in UV-irradiated *E. coli* infected and uninfected with T₂ phage. *E. coli* grown in tris-glucose medium¹ were irradiated with a UV dose, which gave a survival of about 10^{-7} (as determined by colony count) and reduced the protein synthesis to a level less than 10 % of that in non-irradiated cells. The suspension was then infected with T₂ phage, ³⁵S being added at zero time. (M. NAKAMURA collaborated in this experiment.)

non-antigenic protein fraction formed just after infection. (4) The inability of the infected bacteria to continue a synthesis of bacterial enzymes⁵ and also to induce a formation of adaptive enzyme⁶. These data also suggest that the non-antigenic protein formed just after infection may not be bacterial.

From the above discussion, it is concluded that the non-phage-antigenic protein formed immediately after infection is or at least contains a new kind of protein, which is different from both bacterial and phage-specific protein, including phage-precursor protein, but nevertheless is characteristic of phage infection. It does not seem unreasonable, therefore, to speculate that the phage infection first induces in bacteria the formation of a new protein (or ribonucleoprotein**) or proteins and this protein or proteins then plays an important but yet unknown role in producing phage-specific protein and DNA. In any case it is very important and interesting at the present time to investigate the true nature of the non-phage-antigenic protein formed immediately after infection, from both the biochemical and genetical points of view.

Institute of Science and Technology, University of Tokyo, Tokyo (Japan)

ITARU WATANABE

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** Studies on the ribonucleic acid formed just after infection are also in progress in our laboratory and the results will be published elsewhere.