## Phage DNA dynamics and the Correlation with Cell Fate

## Lanying Zeng<sup>1</sup>

Short Abstract — Bacteriophage lambda starts its infection cycle by injecting its DNA into *E. coli*. Upon infection, *E. coli* chooses to follow the lytic pathway – around 100 new phages are released outside the cell, or lysogenic pathway – phage DNA is integrated into the host genome. Phage DNA plays a significant role in both pathways. Here, we apply a new technique to follow the spatiotemporal dynamics of phage DNA movements inside the cell and correlate it with phage infection site and the resulting cell fate under the fluorescence microscope at the level of single cell, single phage and single phage DNA.

*Keywords* — Single cell, single phage, single phage DNA, fluorescently labeled phage, spatiotemporal dynamics of phage DNA, cell fate.

THE post-infection decision process of lytic and I lysogenic pathways serves as a paradigm for an environmentally-regulated genetic switch. The number of infecting phages per cell (known as "Multiplicity of Infection", or MOI) and cell size were shown to be the major parameters to control the cell fate through bulk assays and single-cell studies under the microscope [1-3]. Furthermore, phages (or the injected phage DNAs) seem to act as individual voters, and only when all infecting phages unanimously vote for lysogeny, the cell can be lysogenized [3]. In the lytic pathway, phage DNA replicates with  $\theta$  mode and switches to  $\sigma$  mode before packaging into phage head. About 100 new progeny phages are then released into the environment after cell lysis. In the lysogenic pathway, phage DNA is integrated into the host genome, and replicates along with the host. The spatiotemporal information of the phage DNA either for replication or integration into the host may play an important role in the lysis/lysogeny decision making. Here, we would like to characterize the spatiotemporal dynamics of phage DNA inside the cell and correlate it with the final cell fate. We follow lambda infection, phage DNA inside the cell and cell fate under the microscope, at the level of single cell, single phage and single phage DNA. Infecting phage and cell fate are detected through our standard microscope protocol [3]. Briefly, fluorescently labeled phage allows us to track the number of infecting phages and the infection site and report the lytic pathway. A fluorescent reporter plasmid is used for the detection of the lysogenic pathway. A new technique by use of fluorescent SegA

fusion protein is developed to detect phage DNA [4]. Preliminary data show that phage DNA exhibits behaviors of both localized motion and motion spanning over the whole cell, consistent with the prediction in the literature [5]. From the spatiotemporal information of phage DNA, we could gain a better understanding of how phage DNA location affects the lysis/lysogeny decision-making, and characterize phage DNA movements during the entire infection cycle (immediately early, delayed early and late stages).

## REFERENCES

- Kourilsky P (1973) Lysogenization by bacteriophage lambda I. multiple infection and the lysogenic response. *Mol Gen Genet* 122, 183-195.
- [2] St-Pierre F, Endy D (2008) Determination of cell fate selection during phage lambda infection. PNAS 105, 20705-20710.
- [3] Zeng L, Skinner SO, Zong C, Sippy J, Feiss M, Golding I (2010). Decision making at a subcellular level determines the outcome of the bacteriophage infection. *Cell* 141, 682-691.
- [4] Babic A, Lindner AB, Vulic M, Stewart EJ, Radman M (2008) Direct visualization of horizontal gene transfer. *Science* 319, 1533-1536.
- [5] Parry BR, Surovtsev IV, Cabeen MT, O'Hern CS, Dufresne ER, Jacobs-Wagner C (2014) The bacterial cytoplasm has glass=like properties and is fluidized by metabolic activity. *Cell* 156, 183-194.

<sup>&</sup>lt;sup>1</sup>Department of Biochemistry and Biophysics, Center for Phage Technology, Texas A&M University. E-mail: lzeng@tamu.edu