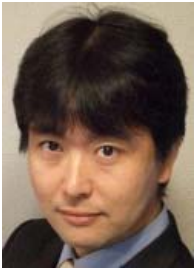


**【Grant-in-Aid for Scientific Research(S)】**  
**Biological Sciences (Biology)**



Title of Project : **Artificial symbiotic system between an insect and *Escherichia coli* toward molecular genetics and experimental evolutionary biology**

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Research Area : Basic biology

Keyword : Experimental evolution, comparative genomics, functional evolution

**【Purpose and Background of the Research】**

Symbiotic associations with microorganisms are ubiquitously found in diverse animals, plants and other organisms, which often play important biological roles for them. How such sophisticated symbiotic systems have been established is an important issue of evolutionary biology.

Recently we found that, in the brown-winged green bugs *Plautia stali* (Hemiptera: Pentatomidae), gut symbiotic bacteria are essential for host's growth and reproduction, and exhibit conspicuous polymorphism within and between host's natural populations. Furthermore, by replacing the original symbionts with *Escherichia coli*, we successfully established an artificial symbiotic system, wherein *E. coli* is normally localized *in vivo*, vertically transmitted through host generations, maintainable in the laboratory, and amenable to experimental and genetic manipulations (Figure 1).

In this project, by making use of molecular genetic, genomic, and experimental evolutionary approaches to this model symbiotic system, we are aiming at understanding of the processes and mechanisms underlying the evolution of symbiosis in unprecedented depth and breadth.

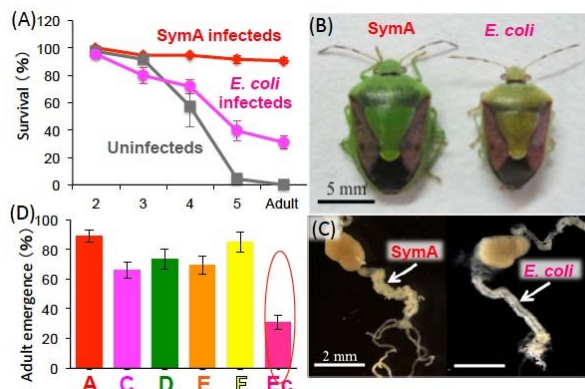


Figure 1 The brown-winged green bug *Plautia stali*. (A) Survival curves of symbiont A-infected, *E. coli*-infected and uninfected insects. (B) External appearance and (C) dissected midgut symbiotic organ of symbiont A-infected and *E. coli*-infected insects. (D) Adult emergence rates of symbiont A-F-infected insects and *E. coli*-infected insects.

**【Research Methods】**

- (1) Complete genome sequencing of 6 symbionts of *P. stali*.
- (2) Establishment of techniques for genetic transformation and gene disruption in the symbionts of *P. stali*.
- (3) Comprehensive isolation of symbiosis-related symbiont genomic regions by screening of *E. coli*-infected insects transformed by fosmid genomic libraries of the symbionts.
- (4) Characterization and functional analysis of symbiosis-related symbiont genes.
- (5) Comparative analysis of symbiosis-related genes of different symbiotic bacteria.
- (6) Experimental evolutionary analysis of *P. stali*-*E. coli* artificial symbiotic association.

**【Expected Research Achievements and Scientific Significance】**

By making use of the sophisticated molecular genetic, genomic and transcriptomic tools and resources of *E. coli* accumulated for decades, we expect that this project will provide unprecedented insights into the molecular mechanisms underpinning the symbiosis. Furthermore, the unique experimental evolutionary approaches to the insect-*E. coli* symbiosis would reveal real-time dynamics and detailed process of host-symbiont co-evolution.

**【Publications Relevant to the Project】**

- Kikuchi Y., Hosokawa T., Fukatsu T. (2008) Diversity of bacterial symbiosis in stinkbugs. In: *Microb. Ecol. Res. Trends* (ed. T. V. Dijk), pp. 39-63. Nova Science Publishers Inc., N. Y.

**【Term of Project】** FY2013-2017

**【Budget Allocation】** 104,100 Thousand Yen

**【Homepage Address and Other Contact Information】**

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