## LABORATORY OF EVOLUTIONARY GENOMICS



Professor: SHIGENOBU, Shuji

Postdoctoral Fellow:	KOBAYASHI, Yuuki
JSPS Postdoctoral Fellow:	NOZAKI, Tomonari
SOKENDAI Graduate Student:	YORIMOTO, Shunta
	TAN, Kathrine
Visiting Scientist:	CHUNG, Chen-yo
Technical Assistant:	SUZUKI, Miyuzu

## **Symbiogenomics**

"Nothing, it seems, exists except as part of a network of interactions." (Gilbert & Epel, 2008)

Every creature on Earth exists among a network of various biological interactions. For example, many multicellular organisms, including humans, harbor symbiotic bacteria in their bodies. Some of them provide their hosts with essential nutrients deficient in the host's diet and others digest foods that are indigestible by the host alone. Despite numerous examples of symbioses and its intriguing outcomes, the genetic and molecular basis underlying these interactions remains elusive. The goal of our group is to establish a new interdisciplinary science known as "Symbiogenomics", where we aim to understand the network of biological interactions at the molecular and genetic level. To this end, we take advantage of state-of-the-art genomics, such as next-generation sequencing technologies and CRISPR-Cas9 genome editing.

## Genomic revelations of a mutualism: the pea aphid and its obligate bacterial symbiont

Aphid species bear intracellular symbiotic bacteria in the cytoplasm of bacteriocytes, which are specialized cells for harboring said bacteria. This mutualism is so obligate that neither can reproduce independently. The genome sequence of the pea aphid, Acyrthosiphon pisum, in consort with that of bacterial symbiont Buchnera aphidicola illustrates the remarkable interdependency between these two organisms (IAGC, PLOS Biol. 2010; Shigenobu et al., Nature. 2000). The genetic capacities of the pea aphid and the symbiont for amino acid biosynthesis are complementary. Genome analysis revealed that the pea aphid has undergone characteristic gene losses and duplications. The IMB antibacterial immune pathway is missing several critical genes, which might account for the evolutionary success of aphids in obtaining beneficial symbionts. Lineage-specific gene duplications have occurred in genes over a broad range of functional categories, which include signaling pathways, miRNA machinery, chromatin modification and mitosis. The importance of these duplications for symbiosis remains to be determined. We found several instances of lateral gene transfer from bacteria to the pea aphid genome. Some of them are highly expressed in bacteriocytes.

We recently discovered a novel class of genes in the pea aphid genome that encode small cysteine-rich proteins with secretion signals that are expressed exclusively in the bacteriocytes of the pea aphid, and named these bacteriocytespecific cysteine-rich proteins (BCR) (Shigenobu & Stern, Proc. R. Soc. B 2013). The BCR mRNAs are first expressed at a developmental time point coinciding with the incorporation of symbionts strictly in the cells that contribute to the bacteriocyte, and this bacteriocyte-specific expression is maintained throughout the aphid's life. Furthermore, some BCRs showed antibiotic activity (Uchi et al., Microbes. Environ. 2019; This paper was selected as the "Most Valuable Paper of the Year 2019" in the journal, Microbes and Environments). These results suggest that BCRs act within bacteriocytes to mediate the symbiosis with bacterial symbionts, which is reminiscent of the cysteine-rich secreted proteins of leguminous plants that also regulate endosymbionts. Employment of small cysteine-rich peptides may be a common tactic of host eukaryotes to manipulate bacterial symbionts.



Figure 1. Pea aphids and the bacterial symbiont, *Buchnera*. Adult aphids (Left). A developing viviparous embryo which symbionts are infecting (Right). Scale bar = 20 um.

## **Publication List**

[Original papers]

- Cai, H., Li, Q., Fang, X., Li, J., Curtis, N.E., Altenburger, A., Shibata, T., Feng, M., Maeda, T., Schwartz, J.A., Shigenobu, S., Lundholm, N., Nishiyama, T., Yang, H., Hasebe, M., Li, S., Pierce, S.K., and and Wang, J. (2019). Data descriptor: A draft genome assembly of the solar-powered sea slug *Elysia chlorotica*. Sci. Data 6, 190022. doi: 10.1038/sdata.2019.22
- Hojo, M., Shigenobu, S., Maekawa, K., Miura, T., and Tokuda, G. (2019). Duplication and soldier-specific expression of geranylgeranyl diphosphate synthase genes in a nasute termite *Nasutitermes takasagoensis*. Insect Biochem. Mol. Biol. *111*, 103177. doi: 10.1016/j. ibmb.2019.103177
- Kutsukake, M., Moriyama, M., Shigenobu, S., Meng, X.-Y., Nikoh, N., Noda, C., Kobayashi, S., and Fukatsu, T. (2019). Exaggeration and cooption of innate immunity for social defense. Proc. Natl. Acad. Sci. USA 116, 8950-8959. doi: 10.1073/pnas.1900917116
- Matsunami, M., Suzuki, M., Haramoto, Y., Fukui, A., Inoue, T., Yamaguchi, K., Uchiyama, I., Mori, K., Tashiro, K., Ito, Y., Takeuchi, T., Suzuki, K.-I.T., Agata, K., Shigenobu, S., and Hayashi, T. (2019). A comprehensive reference transcriptome resource for the Iberian ribbed newt *Pleurodeles waltl*, an emerging model for developmental and regeneration biology. DNA Res. 26, 217-229. doi: 10.1093/dnares/ dsz003
- Morita, S., Ando, T., Maeno, A., Mizutani, T., Mase, M., Shigenobu, S., and Niimi, T. (2019). Precise staging of beetle horn formation in *Trypoxylus dichotomus* reveals the pleiotropic roles of doublesex

depending on the spatiotemporal developmental contexts. PLoS Genet. 15, e1008063. doi: 10.1371/journal.pgen.1008063

- Ohbayashi, T., Futahashi, R., Terashima, M., Barrière, Q., Lamouche, F., Takeshita, K., Meng, X.-Y., Mitani, Y., Sone, T., Shigenobu, S., Fukatsu, T., Mergaert, P., and Kikuchi, Y. (2019). Comparative cytology, physiology and transcriptomics of *Burkholderia insecticola* in symbiosis with the bean bug *Riptortus pedestris* and in culture. ISME J. *13*, 1469-1483. doi: 10.1038/s41396-019-0361-8
- Sekii, K., Yorimoto, S., Okamoto, H., Nagao, N., Maezawa, T., Matsui, Y., Yamaguchi, K., Furukawa, R., Shigenobu, S., and Kobayashi, K. (2019). Transcriptomic analysis reveals differences in the regulation of amino acid metabolism in asexual and sexual planarians. Sci. Rep. 9, 6132. doi: 10.1038/s41598-019-42025-z
- Sugime, Y., Oguchi, K., Gotoh, H., Hayashi, Y., Matsunami, M., Shigenobu, S., Koshikawa, S., and Miura, T. (2019). Termite soldier mandibles are elongated by dachshund under hormonal and hox gene controls. Development *146*, dev171942. doi: 10.1242/dev.171942
- Uchi, N., Fukudome, M., Nozaki, N., Suzuki, M., Osuki, K.-I., Shigenobu, S., and Uchiumi, T. (2019). Antimicrobial activities of cysteine-rich peptides specific to bacteriocytes of the pea aphid *Acyrthosiphon pisum*. Microbes Environ. 34, 155-160. doi: 10.1264/ jsme2.me18148
- Yaguchi, H., Suzuki, R., Matsunami, M., Shigenobu, S., and Maekawa, K. (2019). Transcriptomic changes during caste development through social interactions in the termite *Zootermopsis nevadensis*. Ecol. Evol. 9, 3446-3456. doi: 10.1002/ece3.4976