

FY2021 Annual Report (April 2021-March 2022)

Molecular Genetics Unit

Professor Daniel Rokhsar (Adjunct; Visiting)

Abstract

Genomic perspective on the evolution of plants and animals

The comparative study of genomes provides a window into the origin and evolution of plant and animal diversity. Contemporary genomes evolved by descent with modification from earlier sequences, accumulating both neutral and adaptive mutations, with varying genomic impact ranging from small changes in sequence to radical changes in chromosome number and gene content. We apply the general comparative principle: features that are shared by two or more genomes were either present in their common ancestor, or evolved convergently.

This principle allows us, for example, to infer the gene content and even genome structure of early vertebrate by comparing genomes of appropriately chosen living species, and to discern macro-evolutionary trends. Similarly, analysis of genetic variation within individual species or between closely related populations sheds light on the evolutionary mechanisms underlying the origin and diversification of new species. The dynamic nature of genomes, including the waxing and waning of transposable elements that generate complex repetitive patterns, also provide opportunities for inferring past events that shaped the diversity of life.

1. Staff

- Dr. Daria Gavriouchkina, postdoctoral researcher
- Dr. Chikatoshi Sugimoto, postdoctoral researcher
- Ms. Lin Zhang, technician
- Dr. Ferdinand Marletaz, visiting researcher
- Ms. Kiyoko Yamada, research unit administrator

2. Collaborations

2.1 Theme: Origins and diversity of mandarin citrus

- Type of collaboration: Collaboration
- Researchers
 - Prof. Fred Gmitter (University of Florida, USA)
 - Dr. Albert Wu (DOE Joint Genome Institute, USA)
 - Dr. Hideyasu Kinjyo (Okinawa)
 - Dr. Fumimasa Musebe (Okinawa Prefectural Agricultural Research Center)
 - Prof. Manuel Talon (Instituto Valenciano de Investigaciones Agrarias, Spain)
 - Prof. Masashi Yamamoto (Kagoshima University, Japan)

2.2 Theme: The evolution of metazoan genomes

- Type of collaboration: Collaboration

- Researchers
 - Prof. Jeramiah Smith (U. Kentucky, USA)
 - Prof. Oleg Simakov (University of Vienna, Austria)
 - Prof. Richard E. Green (University of California, Santa Cruz, USA)
 - Prof. Chris Lowe (Hopkins Marine Station, Stanford University, USA)
 - Prof. Rob Steele (University of California, Irvine, USA)
 - Dr. Steven Haddock (Monterey Bay Aquarium Research Institute, USA)
 - Prof. Charles Davis (Ludwig Maximilians University, Munich, Germany)

2.3 Theme: evolution and diversity of cephalopods

- Type of collaboration: Collaboration
- Researchers
 - Prof. Oleg Simakov (University of Vienna, Austria)
 - Dr. Carrie Albertin (Marine Biological Laboratory, Woods Hole)
 - Prof. Cliff Ragsdale (University of Chicago, USA)
 - Dr. Joshua Rosenthal (Marine Biological Laboratory, Woods Hole)
 - Prof. Gustavo Sanchez-Silva (Hiroshima University, Japan)

3. Activities and Findings

3.1 Evolution and diversity of citrus

The origin and dispersal of cultivated and wild mandarin and related citrus are poorly understood, in part due to their propensity for hybridization and asexual reproduction by nucellar embryony (a form of apomixis). We have used genomic, phylogenetic and biogeographic analyses of diverse citrus germplasms to develop a new evolutionary framework for the genus *Citrus* that accounts for primary citrus species and their role in producing admixed or hybrid taxa. Recent analyses of east Asian mandarins revealed a previously unrecognized wild sexual species native to the Ryukyu Islands, which we named *Citrus ryukyuensis*. With this new species the taxonomic complexity of east Asian mandarins collapses to a satisfying simplicity, accounting for shiikuwasha, tachibana, and other traditional Ryukyuan types as hybrid species that reproduce clonally by seed via nucellar embryony. We traced the origin of citrus apomixis alleles to mangshanyehu wild mandarins, which played a central role in citrus domestication via adaptive wild introgression. Our results provide a coherent biogeographic framework for understanding the diversity and domestication of mandarin-type citrus through speciation, admixture, and rapid diffusion by apomictic reproduction.

3.2 The evolution of metazoan genomes

We developed new methods for analyzing genome evolution at the chromosome scale and are using these methods to explore animal diversity. Comparing chromosome-scale genome sequences of diverse bilaterians, cnidarians, and sponges we find extensive conservation of synteny (that is, gene linkage without regard to collinearity). These patterns of conservation allow us to reconstruct ancestral chromosomes across major animal groups, and the transitions between them. Comparisons among diverse metazoans show that the past 500 million years of animal chromosome evolution have been dominated by arm-scale fusions and fissions as well as the previously unappreciated process of fusion-with-mixing. Fusion-with-mixing is an irreversible process in which genes from two ancestral syntenic groups are combined and intermingled on a single chromosome. This rare and irreversible process is a useful phylogenetic character that provides a

new method for establishing clades that share one or more such fusions-with-mixing. For example, we find that all spiralian share four such chromosomal changes; *Trichoplax* and cnidarians share a specific mixing that strongly suggests that placozoans and cnidarians form a clade to the exclusion of other animal groups. Remarkably, we also find evidence of conserved synteny between metazoans and related unicellular eukaryotes, suggesting a very deep ancestry for certain gene linkages. These findings extend our previous studies focused on the evolution of vertebrate chromosomes, in which we used fusions-with-mixing to prove that jawed vertebrates share a common allotetraploid history that is not shared with jawless vertebrates. We continue to apply these methods to other animal and non-animal groups.

3.3 Evolution and diversity of cephalopods

Our unit has a long-standing interest in using genomics to study the nature and diversity of cephalopods. Bobtail and bottletail squid are small cephalopods with striking anti-predatory defense mechanisms, bioluminescence and complex morphology that inhabit nekto-benthic and pelagic environment around the world's oceans. To illuminate the evolution and diversification of these animals we used shallow genome sequencing of thirty-two samples to estimate evolutionary relationships and divergence times. Phylogenetic analysis confirmed the monophyly (evolutionary coherences) of multiple groups within the bobtails and bottletails, and allowed us to infer that the ancestor of the sepioline bobtails possessed a bilobed light organ with bacteriogenic luminescence. Sepioline radiated in the Late Cretaceous and split into distinct Indo-Pacific and Atlantic-Mediterranean lineages when the Tethys Sea closed at the end of the Eocene and beginning of the Oligocene. We are currently working on several new cephalopod genomes, including reference quality genomes for the longfin inshore squid famous for its giant axon and for a Japanese bobtail squid that is an emerging model system for cephalopod biology. Ongoing studies include characterization of life history and culture conditions of Japanese bobtail squid, regulation of RNA editing, and the structure and organization of the central nervous system.

4. Publications

4.1 Journals

1. Simakov, O., Bredeson, J., Berkoff, K., Marletaz, F., Mitros, T., Schultz, D.T., O'Connell, B.L., Dear, P., Martinez, D.E., Steele, R.E., Green, R.E., David, C.N., Rokhsar, D.S. 2022. Deeply conserved synteny and the evolution of metazoan chromosomes. *Sci Adv.* 8(5):eabi5884. doi: 10.1126/sciadv.abi5884.
2. Sanchez, G., Simakov, O., Rokhsar, D.S. 2022. Beyond "living fossils": Can comparative genomics finally reveal novelty? *Mol. Ecol. Resour.* 22(1):9-11. doi: 10.1111/1755-0998.134881.
3. Miryeganeh, M., Marlétaz, F., Gavriouchkina, D., Saze H. 2021. De novo genome assembly and in natura epigenomics reveal salinity-induced DNA methylation in the mangrove tree *Bruguiera gymnorhiza*. *New Phytol.* 233(5):2094-2110. doi: 10.1111/nph.17738.
4. Wu, G.A., Sugimoto, C., Kinjo, H., Azama, C., Mitsube, F., Talon, M., Gmitter, F.G. Jr., Rokhsar, D.S. 2021. Diversification of mandarin citrus by hybrid speciation and apomixis. *Nat. Commun.* 12:4377. doi: 10.1038/s41467-021-24653-0.
5. Sanchez, G., Fernández-Álvarez, F.Á., Taite, M., Sugimoto, C., Jolly, J., Simakov, O., Marlétaz, F., Allcock, L., Rokhsar, D.S. 2021. Phylogenomics illuminates the evolution of bobtail and bottletail squid (order Sepiolida). *Commun. Biol.* 4:819. doi: 10.1038/s42003-021-02348-y.

6. Xu, F., Marlétaz, F., Gavriouchkina, D., Liu, X., Sauka-Spengler, T., Zhang, G., Holland, P. W. H. 2021. Evidence from oyster suggests an ancient role for Pdx in regulating insulin gene expression in animals. *Nat. Commun.* 12(1):3117. doi: 10.1038/s41467-021-23216-7.

4.2 Books and other one-time publications

Nothing to report

4.3 Oral and Poster Presentations

Nothing to report

5. Intellectual Property Rights and Other Specific Achievements

Nothing to report

6. Meetings and Events

Nothing to report

7. Other

Nothing to report