Evolution of immune gene interaction in Zurich

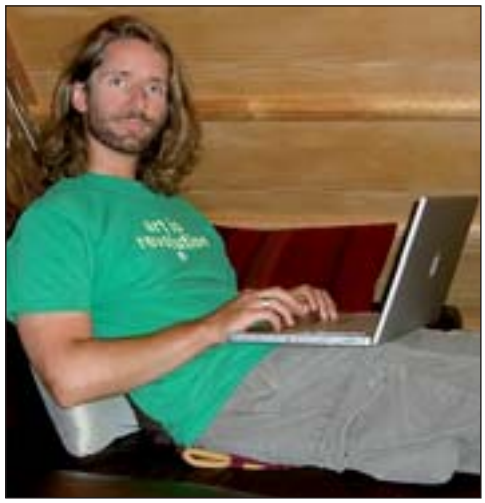
Better Be Clustered

One-author papers have become a rarity in life science research. By applying bioinformatical data mining to reveal the genomic and recombinatorial patterns of immune genes Mathias Wegner from Zurich has recently produced one.

From time to time, scientific publications may leave the critical reader feeling like Alice in Wonderland. Strange questions addressed with equally strange methods, sometimes leading to very peculiar outcomes, could paint a picture of a nonsensical fantasy world of science, where all life's existing rules are displaced. Well, this isn't intended to become a discussion of incredible science. We all believe that science is sometimes a world of marvels. We don't need any any assistance from Alice to tell us that.

Marvellous evolution

Nevertheless, her creator Lewis Carroll, has had an important – and very serious – influence on science, especially in evolution and population genetics research. The Red Queen, a character of the Wonderland sequel *Through the Looking-Glass*, gave her name to an evolutionary hypothesis stating that continuing development is needed for an evolutionary system in order to maintain its fitness relative to the systems with which it is co-evolving. Proposed in the early 70s by Leigh Van Valen, the Red Queen hypothesis is intended to describe appropriately the permanent evolutionary arms race between competing species.



Mathias Wegner demonstrates how to achieve a one-author paper.

"For instance, regarding the co-evolution of parasites and their hosts, changes on both sides presumably lead to the maintenance of diversity, in accordance with the hypothesis principle," says K. Mathias Wegner from the ETH in Zurich, Switzerland. In the Unit of Experimental Ecology of the Institute for Integrative Biology, he studies evolutionary processes and their genetic and molecular backgrounds, especially those of the immune system, using invertebrate model systems. "If the Red Queen hypothesis holds for the evolution of the immune system, the mutual adaptation on the thus generated selection pressure on both sides should leave marks in the genome." How those molecular footprints look and what they reveal about evolution is of interest to Mathias Wegner.

The immune system, which has to react rapidly to a great variety of pathogens attacking the organism, is quite renowned for its diversity. This is achieved, for instance, through the polymorphism of the genes involved in the immune response as well as their genetic recombination. In addition, enabling such complex processes like the immune response to work effectively, the interplay of the various genes involved has to be coordinated.

"For *Drosophila melanogaster*, a large proportion of immune genes interact with each other in a way of epistasis," states Mathias Wegner, meaning the action of one gene is modified by another. Where positive epistasis occurs, the simultaneous presence of two interacting alleles leads to more fitness of the genotype than if the effects of each one alone were summarised.

"In this case, it makes sense to have the genes connected, to assure that their combination is not broken up," explains Wegner. This was also found for a number of *Drosophila* immune receptor genes: they display a high level of linkage disequilibrium since due to their association they frequently occur in combination.

"Generally, the order of genes in the eukaryotic genome is not random but rather organised in clusters," says Mathias Wegner. Those genes are also often functionally related. Their concentration in clusters results in expression advantages, as co-regulated genes can be transcribed together more efficiently when they are localised close to one another. The consequence is a minimal recombination rate of these genes.

On the other hand, during antagonistic host-parasite co-evolution, as soon as a parasite has adapted to a special immunological gene pattern, the breaking up of genetic connections would be favourable to allow a re-puzzling of the genes in order to achieve optimised connections for the newly evolved requirements. "Then, a higher recombination rate between epistatically interacting immune genes is of advantage," according to Mathias Wegner. "The genes should be spread out in the genome and clustering should be minimal." In other words, co-evolution of parasites and hosts would select for higher recombination rates.

The "immunome" organisation

Due to theoretical considerations, the real world is somewhere in between: they predict an optimal recombination rate for gene-loci subjected to selection pressure during co-evolution rather than minimal or maximal.

In order to see how clustering of immune genes on the one hand and their recombination on the other hand engage, Mathias Wegner recently conducted a data mining analysis on the genome of *Drosophila melanogaster* (*PLoS ONE* 3(7):e2835). Being well described and functionally characterised, it presents itself as a good system for such a theoretical and bio-mathematical project.

The biologist studied the spatial and recombinatorial patterns of the genes in the immune system, termed the immunome. "I particularly wanted to discriminate patterns of neutral evolution with simple gene duplications from those resulting from selection for spatial immunome organisation, addressing transcriptional regulation and co-evolution in particular."

Localising a set of 207 immune genes known from recent comparative studies

in the fruit fly genome, Mathias Wegner found that they were strongly concentrated on chromosome 2, thus showing a 'between chromosome clustering'. The other chromosomes were significantly deficient in immune genes compared to a calculated random distribution. Also within the chromosome he found the immune genes to be grouped into a total of 14 clusters. "As I wanted to take recombination into account in this analysis, I studied the clustering within using a sliding window of a size of 2 centimorgan (cM) and shifted it in 1cM steps along the chromosome," the 34-year old describes. "In each window, I counted the number of immune genes and compared that to a calculated random distribution of the genes."

Beyond the primary subject

Additionally, Wegner determined the local recombination rates for the different functional classes of immune genes involved in recognition of the pathogen, signalling or as an effector of defence. He thus identified that signalling genes are less often recombined – not very surprising as they are relatively conserved and also needed in signalling pathways other than in the immune response. In contrast, he found the clusters of immune genes directly interacting with the pathogen, e.g. recognition and effector genes, to be located in regions with high recombination rates. Since higher recombination rates increase the likelihood of fixing beneficial mutations, the direct interaction between host and parasite molecules might have increased selection for cluster formation in regions of higher recombination rates for these classes of genes, surmises the biologist.

Wegner found the clustered effector genes to be transcribed very fast in the beginning of the immune response, indicating that, for those, gene regulation may be the major motor for cluster formation. "However, this can only tell little about the relative orientation of the clusters to each other on the chromosome," he points out. "Possibly, this was arranged through epistasis between genes involved in host parasite interactions," thinks Wegner. Selection for changes in linkage disequilibria and for optimal recombination rates as a consequence of antagonistic, host-parasite co-evolution could potentially explain the non-random order of clusters along a chromosome. This means that a cluster orientation with an optimised distance between them would survive over time. "Besides the idea of transcriptional advantages in gene clusters, this advances another proposal of how genome architecture is organised, not only in local gene clusters but especially on the next hierarchical level of chromosomes," Mathias Wegner summarises.

Such theoretical work using biomathematical tools is not the biologist's usual primary subject. Nor is *Drosophila melanogaster* the main model organism he works on. "I usually do experimental lab work focussing on evolutionary questions and modelling host-parasite co-evolution," Wegner admits. He generates different populations, crossbreeds and infects them with parasites so he may study the effects of the genetic pattern on the gene interaction. "How positive epistasis can switch to negative epistasis over time when hosts and parasites co-evolve, is what we want to understand in detail," Wegner explains.

During his PhD at the Max-Planck-Institute for Evolutionary Biology, formerly the MPI for Limnology, in Plön, the biologist, born on the North Frisian Island of Sylt, studied polymorphic major histocompatibility complex (MHC) genes of sticklebacks and the evolutionary effects of parasite infection. "Those fishes are very good models for the vertebrate system, however, due to their relatively long reproduction time, they cannot compete against insects, when you want to study scientific principles," Mathias Wegner states. Thus, having moved to the ETH in Zurich in April 2005, he turned to his current easy-to-handle model organism, the red flour beetle *Tribolium castaneum*, which he infects with his natural microsporidian parasite *Nosema whitei* to study the dynamics of co-evolutionary processes.

Fiddling until they fit

However, to some extent, Wegner wants to continue with bioinformatics. "I like this kind of work, designing algorithms and fiddling about them until they fit," he says. "You can generate quite an amount of data in relatively short time. And a pleasant thing – you can do it anytime alone, for instance at home on your couch," he adds, laughing. One reason why he is the single author of the paper mentioned.

In future, he wants to use bioinformatic tools mainly as a device, for example, to select interesting candidate genes he would like to analyse further in *Tribolium*. Acquiring more and more insight into the molecular evolutionary processes of the immune system in this animal will surely keep him busy for the foreseeable future.

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