

Bericht über den 1. Workshop der Gen-AG Evolutionsgenetik

Prof. Karl Schmid, Universität Hohenheim

Der erste Workshop der Gen-AG fand vom 28. September bis zum 1. Oktober 2010 in der Evangelischen Jugend-, Freizeit- und Bildungsstätte in Plön statt. Die Hauptorganisatoren waren Dr. Bettina Harr, MPI für Evolutionsbiologie, Plön und Prof. Dr. Karl Schmid, Universität Hohenheim. Dazu wurde die Vorbereitung noch von Emilie Hardouin, Weini Huang und Inka Montero, die Doktoranden oder Postdocs am MPI Plön sind, unterstützt.

Der Workshop wurde über verschiedene Kanäle annonciert, wie der evoldir Mailingliste, über Biospektrum, sowie durch direktes Anmailen von evolutionsgenetisch orientierten Arbeitsgruppen. Die Informationen und Anmeldungen wurden über die Webseite (<http://web.evolbio.mpg.de/~harr/home.html>) abgewickelt.

Inklusive der eingeladenen Sprecher waren 32 Teilnehmer in Plön, darunter acht vom ortsansässigen MPI. Bemerkenswert war, dass zwei Teilnehmer aus Italien und eine Teilnehmerin aus Griechenland kamen. Der Workshop war in erster Linie für Doktoranden ausgeschrieben, und der Anteil der Doktoranden war dementsprechend sehr hoch.

Wir hatten drei Sprecher eingeladen, die restlichen Vorträge wurden von den Teilnehmern bestritten. Das Workshop-Programm ist dem Bericht beigefügt. Die Diskussionen waren sehr rege und setzten sich auch in den Pausen fort. Unser Eindruck war, dass vor allem die Breite der vertretenen Forschungsrichtungen innerhalb der Evolutionsgenetik von der Theorie bis hin zu angewandten Fragestellungen (Conservation Genetics) zu interessanten und fruchtbaren Diskussionen und Kontakten geführt haben.

Ursprünglich hatten wir auch einen sogenannten 'unconference' Teil, mit spontanen Diskussionen und Workshops geplant, aber dann mangels Feedback nicht durchgeführt. Für das nächste Mal müssen wir uns hier besser vorbereiten, um notfalls eigene Themen auf die Agenda setzen zu können.

Wir hatten mit 50 Teilnehmern geplant, aber angesichts mehrerer anderer Veranstaltungen zum Thema Evolution im deutschsprachigen Raum konnten wir nicht mehr Teilnehmer gewinnen. Obwohl wir Stornierungsgebühren bezahlen mussten, haben wir keine Verluste gemacht, da wir die Zuschüsse der GfG von 2009 und 2010 in Anspruch nehmen konnten.

Da der Workshop auch der Mitgliederwerbung dienen sollte, haben die Möglichkeit einer Rückerstattung mehrmals angesprochen. Wir wissen nicht, wieviele neue Mitglieder sich angemeldet haben, aber wir vermuten, dass der finanzielle Anreiz der GfG beizutreten eher gering ist, weil ja die Workshopgebühr in der Regel vom Budget der Arbeitsgruppe bezahlt wird, wogegen die GfG-Mitgliedschaft privat bezahlt werden muss.

Aus unserer Sicht war der Workshop erfolgreich und ein wichtiger erster Schritt für die Gen-AG. Unser Ziel für die Zukunft ist es, die Sichtbarkeit der Gen-AG noch weiter zu erhöhen und die Zahl der Interessenten und Teilnehmer an den Veranstaltungen zu erhöhen.

Stuttgart, 6. Januar 2011



First Workshop

Gen-AG 'Evolutionary Genetics'

German Society of Genetics (GfG)

Kappelsberg, Plön, Germany

28 September - 1 October 2010

Organizers:

Karl Schmid, University of Hohenheim
Tina Harr, MPI for Evolutionary Biology
Emilie Hardouin, MPI for Evolutionary Biology
Weini Huang, MPI for Evolutionary Biology
Inka Montero, MPI for Evolutionary Biology

Support:



Max-Planck-Institute
for Evolutionary Biology

PROGRAMME

Tuesday, 28. September

12:00	Arrival
16:00 - 18:00	Registration
19:00	Dinner

Wednesday, 29. September

8:45	Welcome Address
9:00	Peter Keightley, University of Edinburgh <i>Estimation mutation and selection parameters</i>
10:30	Mingkun Li, MPI of Evolutionary Anthropology, Leipzig <i>Heteroplasmy in Human MtDNA Genomes</i>
10:50	Sebastian Novak, University of Vienna <i>Evolution in spatially structured environments - the Levene model</i>
11:10	Coffee break
11:40	Bin Wu, MPI for Evolutionary Biology, Plön <i>On the universality of weak selection</i>
12:00	Weini Huang, MPI Plön <i>Mutant games</i>
12:20	Lunch
14:00	Chaitanya Gokhale, MPI of Evolutionary Biology, Plön <i>Playing with genes? An evolutionary game theoretic perspective</i>
14:20	Julia Niehörster, MPI of Molecular Plant Physiology, Golm <i>Plastome-Genome incompatibility in the evening primrose <i>Oenothera</i></i>
14:40	Sariel Hubner, University of Haifa <i>Regulating factors of genetic structure in wild barley: The Barley1K as a model</i>
15:00	Inka Gawenda, University of Hohenheim <i>Towards haplotype-based genome-wide association mapping in crop plants with an application in barley</i>
15:20	Coffee Break
15:50	Torsten Günther, University of Hohenheim <i>Methods for natural selection mapping in plant genomes</i>

- 16:20 Oliver Simon, University of Hohenheim
Studies on the natural variation of the oil content of Arabidopsis thaliana seeds
- 16:50 Thomas Müller, University of Hohenheim
Adaptation of forest trees to climatic change - Diversity of drought responses in Douglas-fir provenances
- 17:10 D. Yahiaoui, Mediterranean Agronomic Institute, Valenzano, Italy
Study on the genetic diversity of Citrus Tristeza Virus (CTV) populations in order to predict eventual disease outbreaks
- 19:00 Dinner
- 20:00 Beer and Snacks

Thursday, 30. September

- 9:00 Hopi Hoekstra, Harvard University
White mice on white sand: The molecular steps to an adaptive peak
- 10:30 Andri Manser, University of Zurich
Polyandry and the decrease of a selfish genetic element in a wild house mouse population
- 10:50 Emilie Hardouin, MPI of Evolutionary Biology, Plön
House mouse colonization patterns on the sub-Antarctic Kerguelen Archipelago
- 11:00 **Coffee break**
- 11:30 Hernan Burbano, MPI of Evolutionary Anthropology, Leipzig
Targeted sequencing of Neandertal DNA: insights into recent human lineage coding and non-coding evolution
- 11:50 Juan Valqui, University of Kiel
Conservation status of the marine otter (Lontra felina) in Peru - Field data and first genetic results
- 12:20 **Lunch**
- 14:00 **Unconference session**
- 16:30 Samer Sermani, University of Bari
Polymorphism in pathogenicity in species of Verticillium phytopathogens
- 17:00 Ahmed Hussien, University of Bari
Studies on pathogenicity characters and its evolutionary dynamics in Fusarium spp. associated to citrus plantation

- 17:20 David Bogumil, University of Düsseldorf
Accelerated substitution rates in Prokaryotes
- 17:50 Ovidiu Popa, University of Düsseldorf
A directed network of recent lateral gene transfer within prokaryotes reveals trends and barriers in gene acquisition
- 18:10 Jennifer Rieger, IFM GEOMAR
How specific is innate immunity? – Experimental resistance evolution in three-spined stickleback
- 18:30 Noémi Erin, MPI for Evolutionary Biology, Plön
Host-parasite G x G interactions in sympatric and allopatric combinations
- 19:00 Dinner
- 20:00 Beer and Snacks

Friday, 1. October

- 9:00 Frank Chan, MPI for Evolutionary Biology, Plön
Adaptive Swarms: a Massively Parallel Approach to Genomics of Adaptation
- 10:30 Jie Cheng, MPI for Evolutionary Biology, Plön
Genetic mapping of hybrid traits in invasive sculpins
- 10:50 Bernd Hermann, IFM GEOMAR Kiel
*Impact of a catastrophic flood event on the population structure of a locally adapted extremophile fish, *Poecilia mexicana**
- 11:10 Coffee break
- 11:30 Emanuella Vogiatzi, Hellenic Centre for Marine Research, Crete
*Phylogeography of the striped red mullet (*Mullus surmuletus* L.) inferred from microsatellite markers and mtDNA.*
- 11:50 Joshka Kaufmann, MPI for Evolutionary Biology, Plön
Post-copulatory reproductive isolation during ongoing ecological speciation in three-spined stickleback
- 12:10 Philip Altrock, MPI for Evolutionary Biology, Plön
Using underdominance to bi-stably transform local populations
- 12:30 Lunch
- 14:00 Departure

ABSTRACTS

(alphabetically sorted)

Using underdominance to bi-stably transform local populations

Philip Altrock

Research Group Evolutionary Theory, Max-Planck-Institute for Evolutionary Biology, Plön

Underdominance refers to natural selection against individuals with a heterozygous genotype. Here, we analyze a single-locus underdominant system of two large local populations that exchange individuals at a certain migration rate. The system can be characterized by fixed points in the joint allele frequency space. We address the conditions under which underdominance can be applied to transform a local population that is receiving wildtype immigrants from another population. In a single population, underdominance has the benefit of complete removal of genetically modified alleles (reversibility) and coexistence is not stable. The two population system that exchanges migrants can result in internal stable states, where coexistence is maintained, but with additional release of wildtype individuals the

system can be reversed to a fully wildtype state. This property is critically controlled by the migration rate. We approximate the critical minimum frequency required to result in a stable population transformation. We also concentrate on the destabilizing effects of fitness and migration rate asymmetry. Practical implications of our results are discussed in the context of utilizing underdominance to genetically modify wild populations. This is of importance especially for genetic pest management strategies, where locally stable and potentially reversible transformations of populations of disease vector species are of interest.

Chaperonin-Dependent Accelerated Substitution Rates in Prokaryotes

David Bogumil

Institute of Botany III, University of Düsseldorf, Germany

Many proteins require the assistance of molecular chaperones in order to fold efficiently. Chaperones are known to mask the effects of mutations that induce misfolding, because they can compensate for the deficiency in spontaneous folding. One of the best studied chaperones is the eubacterial GroEL/GroES system. In *Escherichia coli*, three classes of proteins have been distinguished based on their degree of dependency on GroEL for folding: I) those that do not require GroEL, II) those that require GroEL in a temperature-dependent manner, and III) those that obligately require GroEL for proper folding. The buffering effects of GroEL have so far been observed in experimental regimens, but their effect on genomes during evolution has not been examined. Using 446 sequenced proteobacterial genomes, we have compared the frequency of amino acid replacements among orthologs of 236 proteins corresponding to the three categories of GroEL dependency determined for *E. coli*. Evolutionary rates are significantly correlated with GroEL-dependency upon folding with GroEL-dependency class accounting for up to 84% of the variation in amino acid substitution rates. Greater GroEL-dependency entails increased evolutionary rates with GroEL obligatory proteins (class III) evolving on average up to 15% faster than GroEL partially dependent proteins (class II) and 35% faster than GroEL independent proteins (class I). Moreover, GroEL-dependency class correlations are strictly conserved throughout all proteobacteria surveyed, as is a significant correlation between folding class and codon bias. The results suggest that during evolution, GroEL-dependent folding increases evolutionary rate by buffering the deleterious effects of misfolding-related mutations.

Targeted sequencing of Neandertal DNA: insights into recent human lineage coding and non-coding evolution

Hernán A. Burbano

Department of Evolutionary Genetics, Max Planck of Evolutionary Anthropology, Leipzig, Germany.

Neandertals, who became extinct around 30,000 years ago, are the closest relatives of extant humans. Comparison of Neandertals and present-day human genomes can reveal information about whether genetic changes occurred before or after the population split of modern humans and Neandertals. We used hybridization capture on microarrays to generate large scale sequencing data from a ~49,000-years-old Neandertal specimen from El Sidrón, Spain. We have sequenced two different groups of Neandertal sequences: i. Protein coding positions (~14,000) that have been inferred to have changed on the human lineage since the last common ancestor shared with chimpanzees; ii. Protein non-coding regions (~2,600) that are both highly conserved in vertebrate evolution yet fast-evolving on the human lineage (human accelerated regions). Using the Neandertal sequences in conjunction with present-day humans polymorphism information, we were able to time either how long ago the original mutation occurred or the subsequent fixation event took place. I will present an in depth analysis of both kinds of captured positions.

Host-parasite G x G interactions in sympatric and allopatric combinations

Noémie Erin, Martin Kalbe and Irene Samonte-Padilla
Max-Planck-Institute for Evolutionary Biology, Plön, Germany

Coevolution through host-parasite interaction is a powerful evolutionary force in adaptation process. We aim to investigate the molecular genetic basis of evolving host resistance and parasite virulence by using sympatric (coevolved) and allopatric (non-coevolved) combinations of threespine stickleback (*Gasterosteus aculeatus*) and its specific tapeworm *Schistocephalus solidus*. The strong phenotypic differences observed in crosswise infection experiments using Norwegian and German populations of our host-parasite system suggest different resistance/virulence strategies for the different origin of host/parasite allowing a comparison of differential gene expression of immunological relevant genes in the different combinations on both host and parasite side.

Adaptive Swarms: a Massively Parallel Approach to Genomics of Adaptation

Yingguang Frank Chan^{1,2}, Felicity Jones² and Stickleback Genome Consortium (led by Richard M. Myers³, Federica Di Palma⁴, Kerstin Lindblad-Toh⁴ and David M. Kingsley²)

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Adaptation is widespread in nature. Despite its obvious importance, we know little about how adaptation happens. To identify the genetic changes that underlie adaptation, we chose to study replicated three-spined stickleback populations. Since the retreat of the Pleistocene ice sheet, sticklebacks have colonised novel freshwater environments and undergone a rapid adaptive radiation, often reaching similar adaptive phenotypes independently. I will start by summarising lessons drawn from classical genetic mapping approaches to identify genes controlling adaptive traits in sticklebacks. Then I will discuss a new approach we have developed that 1) systematically identify any loci controlling parallel adaptation via re-use of standing variations; and 2) generate new ecological hypothesis for unknown adaptations. We have generated next-generation sequences for 21 individual sticklebacks from around the world. We have developed methodologies that can identify the genetic substructure among the fish in a genomic region. Besides providing details on the phylogeography of sticklebacks, our approach identifies a large number of candidate regions underlying adaptations. I will focus our discussion on regions that show strong parallel differentiation between marine and freshwater sticklebacks. These regions correspond to regions of elevated divergence between samples taken from opposite ends of a hybrid zone. Deeper population-level sampling recovers diversity patterns consistent with selective sweeps. I will discuss several general principles emerging from the genome-wide pattern, including the possible origin(s) of adaptive alleles; and chromosomal characteristics that favour adaptation via standing variation, with special emphasis on the role of chromosomal inversions.

Genetic mapping of hybrid traits in invasive sculpins

Jie Cheng

Department of Evolutionary Genetics, Max-Planck Institute for Evolutionary Biology, Plön, Germany

This study focuses on a likely case of hybrid speciation in which two fish species (*Cottus rhenanus* and *Cottus perifretum*) have interbred to give rise to a hybrid lineage with a new ecological potential to invade disturbed habitats that were not accessible to their parental species before. Now next-generation sequencing allows the discovery of large numbers of single nucleotide polymorphisms (SNPs) in species where little genomic information was previously available. Here, 454 sequencing was applied to perform a genome-wide screen to identify genetic factors that are associated with the hybrids success. We assembled, de novo, about 350 Mb of non-normalized cDNA 454 data from *C. rhenanus* (river Naaf and river Broel), *C. perifretum* (river Wite Nete and river Laarsebeek) and hybrids (river Sieg). To better allocate the parental origin of the genome regions in the hybrid lineages, our main goals were to gather a large data set of ancestry informative SNP markers whose alleles are reciprocally fixed or nearly fixed in each species. We identified 1,302 ancestry informative SNPs in 27,236 contigs. The resulting sequences were compared by BLAST against the stickleback genome with e -value $< 10^{-6}$, 1000 ancestry informative SNPs have significant hits and be mapped on 21 groups. Inspection of these fragments allowed us to identify candidate loci where particular positions or genomic blocks appeared to be alternatively fixed in parental species. Against the stickleback genome, we also mapped the SNPs from 6 hybrid individuals separately which indicates that there are some regions that have only one parental ancestry. Using likelihood inference method, we estimate that the block size in hybrids might be about 2.5Mb, which is much bigger than former results. Further on, we are going to perform genomic sequencing in order to get more and genome-distributed ancestry informative markers and genetic map candidate loci evenly distributed across the genome to validate our result.

Towards haplotype based genome-wide association mapping in crop plants with an application in barley

Inka Gawenda

Department of Plant Breeding, Seed Science and Population Genetics, University of Hohenheim, Stuttgart, Germany

In the last years, association mapping approaches became a widely used method to map qualitative and quantitative traits in plants. For example, numerous studies for association mapping were conducted in barley. Next to single marker association methods, multipoint association methods which are based on a perfect phylogenetic tree of linked markers were developed (Templeton et al., 2005, Mailund et al., 2006, Besenbacher et al., 2009). To reduce the multiple testing problem, the effect of closely located markers is considered by using neighboring markers or calculating a perfect phylogeny tree. Haplotype-based multipoint methods offer two major advantages: first, they reduce the number of loci to test and thus avoid over-conservative correction for multiple testing; second, these haplotype blocks rather than single markers represent the entire inherited segments. We use simulations to estimate the power of multipoint association methods to compare them with single-marker associations and apply them to yield traits in a collection of German winter barley cultivars.

Playing with genes? An evolutionary game theoretic perspective.

Chaitanya S. Gokhale

Research Group for Evolutionary Theory Max Planck Institute for Evolutionary Biology, Plön, Germany

Traditionally game theory has been used in economics to study conflict scenarios and decision making amongst humans. John Maynard Smith and George Price saw its potential applications in biology and developed Evolutionary Game Theory. Since then Evolutionary game theory has become quite popular amongst behavioral ecologists, sociologists, philosophers and also back amongst economists. Despite other contribution of Maynard-Smith to the field of population genetics, the application of evolutionary game theory in population genetics has been limited. One criticism was that the genetic systems are too complex for the simplicity of evolutionary game theory to handle. Considerable development has since occurred in the field of evolutionary game theory. We show that not only is the traditional evolutionary game theoretical approach capable of capturing simple genetic systems but also the development of multiplayer and multi-strategy game theory can deal with the complicated scenarios e.g. genetic conflict scenarios, yet keeping the essence of evolutionary game theory, simplicity.

Methods for natural selection mapping in plant genomes

Torsten Günther

Department of Plant Breeding, Seed Science and Population Genetics, University of Hohenheim, Stuttgart, Germany

Current next-generation sequencing projects provide sufficiently large datasets for the genome-wide detection of natural selection in many species. Genome-wide scans were able to detect new candidate regions for selection as well as previously identified target genes in humans and other model species. However, unbiased and time-efficient methods are required for the analysis of genome-wide datasets. We present methods to analyze the signature of positive and purifying selection in plant genomes. These methods were tested on simulated datasets under various complex demographic models to assess their power on empirical data. Additionally, we applied the tests to genome-wide SNP and sequence data from *Arabidopsis thaliana*.

House mouse colonization patterns on the sub-Antarctic Kerguelen Archipelago.

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The western house mouse (*Mus musculus domesticus*) is found on most continents and in a wide range of habitats, probably due to an almost exclusively commensal lifestyle. House mice colonized the Kerguelen Archipelago (48°25'-50°S; 68°27'-70°35'E) approximately 200 years ago. These islands are characterized by a typical sub-Antarctic climate and the absence of human settlements, thus the mice have adopted a feral lifestyle and adapted to a tremendously different habitat with cold year-long temperatures and new food resources. We are using population genetic approaches to identify the molecular basis of the major adaptations that enabled the mice to persist and maintain a large population size on Kerguelen. We investigated the colonization history and population structure on the archipelago using mitochondrial and DNA markers on 433 individuals from 15 sites. As expected, Kerguelen mice show very low genetic diversity due to founder effects resulting from recent colonization and geographic isolation. Interestingly, mice from different islands of the archipelago show distinct genetic patterns, suggesting colonization occurred from multiple source populations.

Impact of a catastrophic flood event on the population structure of a locally adapted extremophile fish, *Poecilia mexicana*

Bernd Hermann

IFM GEOMAR, (Leibniz Institute for Marine Sciences), Kiel, Germany

Local adaptation to divergent environmental conditions can promote population genetic differentiation even in the absence of geographic barriers and hence lead to speciation. The process of increasing genetic differentiation between populations can be countered by gene flow, e.g., due to individual translocation caused by catastrophic climatic events. In this work, we compared the genetic population structure of a livebearing fish, *Poecilia mexicana*, in the Cueva del Azufre System at two specific times: Before and after a catastrophic flood event in fall 2007. The Cueva del Azufre system is characterized by the absence or presence of two strong selective agents: darkness and hydrogen sulfide. Using microsatellites as genetic markers, our analyses yielded the same three genetically distinct clusters in both data sets. Each of the three clusters could be assigned to a specific habitat type, depending on its abiotic properties.

Based on the finding that the genetic population structure was maintained even after a major natural disturbance, we infer that strong selection against migrants is operating. Previous studies provide evidence that a combination of both natural and sexual selection is operating. Although genetic structuring between the populations remained constant, we found traces of the flood within the cave-inhabiting population. Small-scale genetic differentiation between the different cave chambers was found before but not after the flood, indicating increased gene flow as a result of individual translocation.

White mice on white sand: the molecular steps to an adaptive peak.

Hopi E. Hoekstra

Dept. of Organismic and Evolutionary Biology, Harvard University

Understanding how organisms adapt to their environments is a great challenge in biology. Recent genomic technologies are enabling us to uncover the molecular details of adaptation, and allowing us to answer fundamental questions, such as: which genes and mutations are involved in adaptive change, and how do these genes interact to produce phenotypic variation? Beyond the mechanistic details, we would also like to know about the evolutionary history of adaptive alleles in nature: where do these alleles come from? how often do the same genes produce convergent phenotypes? In this context, I will present some of our latest results --from both the laboratory and the field -- on the molecular, genetic and developmental changes responsible for adaptation in natural populations of mice.

Mutant games

Weini Huang

Research Group for Evolutionary Theory, Max Planck Institute for Evolutionary Biology, Plön, Germany

The diversity of biological system is maintained by the interaction of mutation, selection and random drift. Even in a haploid and asexual population, polymorphism of alleles is often observed. Mutation brings new alleles all the time, and consequently no allele will stay in a finite population forever. What kind of combination of variant nature power, will lead to a stable polymorphism of alleles? What cause the different allele diversity in different genetic environments, for example, in different locus? We are interested in answering these questions by evolutionary game theory method.

Regulating Factors of Genetic Structure in Wild Barley: The Barley1K as a Model

Sariel Hubner

Institute of Evolution, University of Haifa, Haifa, Israel

Intensive breeding programs for hundreds of years had reduced considerably the genetic variation within available breeding material, causing vulnerable varieties that may not sustain in future environmental conditions. Enrichment of the available breeding gene pool with beneficial alleles originated from natural populations of crops wild ancestors is becoming more attractive due to the allelic richness that evolved in response to environmental pressure. Therefore, an increased interest is noticed in studying the patterns of genetic diversity and its regulation in crops wild ancestors germplasms. In order to investigate genetic diversity in natural populations it is essential to establish a well defined, systematic and consistent infrastructure of wild germplasm that is well representing the species diversity in different environments. Wild barley (*Hordeum spontaneum*) is the direct progenitor of one of the most important crop plants cultivated worldwide and it is abundant in different environments along the Fertile Crescent, where it was domesticated 10,000 years ago.

In this study, 51 wild barley populations were sampled around Israel in a hierarchical sampling mode during the spring of 2007. Altogether, 1020 accessions were collected, representing a wide range of eco-geographical niches. This collection (Barley1K) was fingerprinted using 42 microsatellite (EST-SSR) markers representing all seven chromosomes. According to the genetic and environmental analysis, the sampled sites could be divided into different clusters in accordance with different geographic regions, aridity and temperature gradients. In addition, it was found that historical and recent allelic interchange between the cultivated and wild gene pools is affecting the wild population structure and the ability to detect it.

It is therefore suggested that the genetic make up of wild ancestors populations is regulated mostly by strong environmental and geographic barriers but is also affected by sporadic interchange of gametes between the two sub species.

Studies on pathogenicity characters and its evolutionary dynamics in *Fusarium* spp. associated to citrus plantation

Ahmed Hussien

University of Bari, Italy

Many *Fusarium* spp. have been isolated from citrus plant roots and rhizosphere, and either they were identified as natural inhabitants, secondary invaders, or primary casual agents of diseases.

F. oxysporum and *F. solani* are cosmopolitan soil-borne fungi that are regularly isolated from diseased roots of many plant hosts, although they may not cause the reported disease, they are frequently misidentified as primary causal agents of diseases supported only on their isolation from diseased plants. Considerable variations exist among *Fusarium* spp. strains in terms of physiological, epidemiological, morphological, and in inducing citrus disorders particularly dry root rot. This could allow us to say that not all *Fusarium* strains isolated from infected citrus root, should be seen as causal agents, but most probably, some strains cause the disease, while others work as opportunistic secondary invaders attack only weaken trees, or saprophytic microflora feed on dead organic residues, and mostly morphologically indistinguishable from pathogenic strains.

Recently, mobile pathogenicity chromosomes have been identified in *Fusarium* spp. that can transfer an entire panel of genes required for host compatibility to a new genetic lineage. The correlation between phylogeny, genotype, and pathogenicity phenotypes of *Fusarium* spp. population isolated from citrus rhizosphere can give us more insight knowledge for dynamics of pathogenicity evolution. The pathogenicity is tested through assessment of disease symptoms on artificially inoculated citrus plants, and assessed through quantitative and qualitative analysis of toxins secreted from *Fusarium* strains. Also, we will investigate the polymorphism between non-pathogenic, low, mild, and strong aggressive strains, using AFLP. Meanwhile, phylogeny analysis will be carried out using house-keeping genes sequences (beta tubuline and alfa elongation factor) and more fast evolving markers (like ITS). Then the pathogenicity phenotype of those strains will be correlated with phylogeny analysis requiring extensive usage of powerful bioinformatics tools, example SIMMAP, CONTRAST, BaTs, and Mesquite. From the results of this work, we expect to correlate phylogenetic groups to pathogen aggressiveness, what are the differences between those strains with higher pathogenicity capabilities and low pathogenicity? and ultimately, tracing pathogenicity traits in the phylogeny tree of those strains by molecular markers.

Post-copulatory reproductive isolation during ongoing ecological speciation in three-spined stickleback

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Ecology is now recognized to play a major role in speciation. Reproductive isolation, that is barriers to gene flow, can evolve between populations as a result of ecologically divergent natural and sexual selection. The different forms of reproductive isolation have been well studied on our model species, the three-spined stickleback (*Gasterosteus aculeatus*), from selection against migrants to selection against hybrids. However, knowing that competition between sperm of different males occurs in natural populations as a result of sneaking behavior, it appears important to consider the importance of the reproductive barriers at the gamete level. Here, we examine gamete preference according to habitat in recently diverged subpopulations in northern Germany. Post-copulatory pre-zygotic reproductive isolation will be investigated in two pairs of river-lake populations. The outcome of sperm competition between a sympatric male and males from different populations will be analyzed using neutral markers. We will record sperm concentration and velocities, using C.A.S.A., thus taking into account the factors that might influence fertilization success. We predict a higher fertilization success of sympatric males against males from a different habitat. Such results would demonstrate the role of gamete selection in the process of population divergence and speciation.

Estimating mutation and selection parameters

Peter D. Keightley

Institute of Evolutionary Biology, University of Edinburgh, UK

The mutation rate at the molecular level and the nature of the distribution of fitness effects of new mutations are central questions in evolutionary genetics. In this talk, I describe recent progress in obtaining estimates of these important entities. I describe the application of methodologies that have made it possible to directly estimate the mutation rate per base pair by comparing the genomes of mutation accumulation (MA) lines. I discuss the application of Illumina sequencing and an earlier approach (DHPLC) to measure the mutation rate in *Drosophila melanogaster* MA lines. I then go on to describe Kondrashov and Crow's idea of estimating the genome-wide deleterious mutation rate (U) by combining estimates of the per nucleotide mutation rate with an estimate of the proportion of new mutations in the genome that are deleterious. Applying this approach in *Drosophila* indicates that most mutations in both coding and noncoding DNA are deleterious and that U exceeds 1.4, i.e., each fly has at least 1.4 new deleterious mutations that its parents did not have, that are destined to be eliminated by natural selection. Finally, I describe how the frequency distribution of segregating nucleotide polymorphisms can be used to estimate parameters of the distribution of fitness effects of new mutations, and, by combining this with between-species divergence, to estimate the fraction of adaptive substitutions.

Heteroplasmy in Human MtDNA Genomes

Mingkun Li, Anna Schoenberg, Ivane Nasidze, Mark Stoneking

Department of Evolutionary Genetics, Max Planck Institute for Evolutionary Anthropology, Leipzig, Germany

Heteroplasmy is defined as the presence of more than one type of mtDNA in a cell or tissue. It is known that heteroplasmy exists at some level in healthy individuals and that pathogenic mutations are frequently heteroplasmic. Understanding the basis of heteroplasmy is therefore of both academic and clinical interest.

Although the methodology for detecting heteroplasmy has been improving rapidly, more accurate and efficient methods are needed to accumulate more data, especially for the coding regions, as most previous studies were restricted to the control region. The advent of high-throughput sequencing technologies enables sequencing of complete mtDNA genomes more efficiently, economically, and in a greater depth. Here we sequenced 135 samples from 5 populations on the Illumina GAII platform using a multiplex sequencing protocol for sequencing libraries prepared from two long-range PCR products which span the entire mitochondrial genome. After quality filtering and removal of duplicate reads the average coverage per position is about 80-fold, which via a simulation study should be sufficient to reliably detect heteroplasmic positions with a minor allele frequency greater than 10%.

In order to reduce the false positive rate arising from sequence errors, quality score and strand information were used to develop stringent criteria for calling heteroplasmic positions. Finally, our criteria identified 37 substitution heteroplasmies in 32 individuals. Five individuals possessed two heteroplasmies, and 3 heteroplasmies occurred at the same position in two different individuals. Thirteen heteroplasmies were located in the control region, which is significantly more than expected if heteroplasmies are occurring randomly across the mtDNA genome. We also identified three indel heteroplasmies, one each among three individuals.

The mutational spectrum does not differ between heteroplasmic mutations and polymorphisms in the same individuals, but the relative mutation rate at heteroplasmic mutations is significantly higher than that estimated for all mutable sites in the human mtDNA genome. Moreover, there is also a significant excess of non-synonymous mutations observed among heteroplasmies, compared to polymorphism data from the same individuals. Both mutation-drift and negative selection influence the fate of heteroplasmies to determine the polymorphism spectrum in humans.

Polyandry and the Decrease of a Selfish Genetic Element in a Wild House Mouse Population

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The *t* haplotype is a selfish genetic element that is present in many house mouse populations despite deleterious effects on individuals. By distorting the transmission ratio, *+t* males transmit the *t* haplotype to up to 90% of their offspring. On the other hand, *t/t* individuals perish *in utero*. Theoretical models based on these properties predict a much higher *t* frequency than is actually observed within populations, leading to the *t* paradox. In the present study, the *t* haplotype is examined in an intensively studied wild population near Zurich. We found a significant decrease of the *t* frequency over a time period of 5.5 years that cannot be explained by the effect of transmission ratio distortion and recessive lethals, despite significantly higher life expectancy of *+t* females compared to *+/+* females. Polyandry has been proposed as a general female counterstrategy against the negative fitness consequences of distorters. We investigated the possible influence of polyandry on the *t* system by parameterizing and applying a model to this specific situation. Extensive life history analysis allows us to estimate a generation time of 263 days, which is much higher than usually assumed under natural conditions. Using this and other parameters, we show that polyandry and sex dependent selection can explain the observed *t* dynamics, making it a biologically plausible explanation for low *t* frequencies observed in natural populations in general.

Adaptation of forest trees to climatic change - Diversity of drought responses in Douglas-fir provenances

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The main goal of this project is to understand the response of Douglas-fir provenances to drought in detail. The areas forested with Douglas-firs in Central Europe increase due to their immense growth potential, the impressive technical properties of the wood, the high commercial value of the timber and the yet only small pest and disease problems. Like the areas the necessity of knowledge how the trees will adapt and acclimate to the changing climate conditions - resulting in a significant increase in temperature and more frequent extreme summer drought events - increases.

In this work we aim to identify genes which are involved in the response of Douglas-fir to drought and to identify the differences in the response between different provenances. We have in total 12 cDNA libraries originating from different Douglas-fir provenances, tissues and drought stress conditions. The libraries were sequenced using the 454 technology. After assembling the libraries we search for candidate genes which are present/absent under drought stress conditions. Furthermore we do SNP detection to identify differences in the drought stress response between different provenances at a molecular scope. At a later stage there will be an association study in which the results of this and other projects (regarding phenotypic and metabolic variations) are combined together.

Plastome-Genome incompatibility in the evening primrose *Oenothera*

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The eukaryotic plant cell is a result of co-evolution between the three genetic compartments, the nucleus, the mitochondria and the plastid genome. It becomes particularly apparent, when organelles are exchanged even between closely related species. This often leads to so-called cytoplasmic incompatibilities (CI), which are the result of a disharmonic interaction between the nucleus and the organelle genomes. Recent work has revealed the great importance of CI in evolution. In accordance with the Dobzhansky-Muller model of speciation processes, CI provides a simple mechanism of establishing hybridization barriers.

The model plant *Oenothera* (evening primrose) is an organism perfectly suited to study the molecular mechanisms and the selection forces leading to plastid-mediated CI, so-called plastome-genome incompatibility (PGI). The *Oenothera* combines several genetic features, notably permanent translocation heterozygosity, biparental transmission of plastids, and interfertility of species, which allow an exchange of plastids and haploid chromosomal sets between species.

A systematic exchange of all plastids and nuclear genomes between all *Oenothera* species lead to the identification of three basic nuclear genomes (A, B and C) occurring in homozygous (AA, BB, CC) or stable heterozygous (AB, AC and BC) constitution and five basic genetically distinguishable plastome types (I - V). All haploid nuclear genomes and plastid types are freely combinable in altogether 30 combinations. Only 12 of them are phenotypical green, and of these only seven exist as species. The remaining 18 combinations display PGI to various degrees and can occur naturally as inviable hybrids.

In a pilot study the incompatible combination AB-I was chosen to identify molecular determinants causing PGI in *Oenothera*. This incompatibility builds a strong hybridization barrier between AA-I and AB-II/AB-III species. It appears, that plastome type I is incompatible in the AB background, but the combination AB-II, AB-III, and AB-IV remain green. Sequence comparison of these four plastomes unveils a specific deletion of 148 bp in plastome I, which is not present in plastomes II- IV, turning this deletion to an appealing candidate locus. The deletion affects the divergently operating promoter region between the *psbB* gene, a core subunit of photosystem II and the *clpP* gene, encoding a plastid protease. Physiological and molecular analysis substantiate the relevance of this locus causing PGI in AB-I *Oenothera* plants. These results point to the fact that altered gene regulation of plastid genes may play a role in the microevolution of the genus probably as mechanism of adaptation to changing environmental conditions.

Evolution in spatially structured environments - the Levene model

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Locally varying living conditions are important promoters that bring forward and maintain the genetic variability of populations. In this context, the Levene subdivided population model (Howard Levene, 1953) is the simplest mathematical model which describes the evolution of gene frequencies in a spatially structured population. Individuals are heterozygous and only one locus (with an arbitrary number of alleles) is taken into account to determine a genotype's local fitness in each ecological niche. Migration between demes is assumed to be random; more precisely, offspring is dispersed randomly over all niches according to their relative sizes. From this, one can see directly that in fact we have no structure in the population per se, but consider a well mixed population with locally varying fitness. Furthermore, the model considers selection only, i.e. we ignore effects as mutation or the like. In the field of mathematical population genetics, Levene's model was the first successful attempt to get insight into the mechanisms and consequences of migration and spatial structure.

Despite the equations' relative simplicity the Levene model still poses some open questions. I give a brief introduction into the model and state the existence (and relevance) of a Ljapunov function for these equations. Furthermore I present a finding from my diploma thesis, which shows how many polymorphic equilibria can occur in the Levene model with two alleles only. This gives a good feeling about the variety of dynamical possibilities opening up even for that rather simple and restrictive model.

A directed network of recent lateral gene transfer within prokaryotes reveals trends and barriers in gene acquisition

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Lateral gene transfer (LGT) is an important mechanism of natural variation among prokaryotes, but the extent of genomic exchange among different species and possible barriers to it are still debated. Here we report the use of directed phylogenetic networks that capture both vertical inheritance and recent lateral gene transfer among 657 prokaryotes. Among the 2,148,623 genes in our dataset, 446,854 genes are identified as recent acquisition. This corresponds to an average of 21% recently acquired genes in each genome, with similar proportions observed across different taxonomic groups.

For 7% of the acquired genes a candidate donor gene was identified, among the available genomes by a combination of statistical inference and phylogenetic reconstruction. Directed networks are a well-suited tool to identify, represent and investigate the lateral component of prokaryote genome evolution, which is substantial in magnitude and not tree-like in nature. Directed networks allow us to formulate and test a wide range of hypotheses regarding LGT patterns for mechanisms operating in nature.

How specific is innate immunity? – Experimental resistance evolution in three-spined stickleback

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Host-parasite interactions are highly dynamic and give rise to rapid coevolutionary processes. In this project, we will use artificial selection to investigate specificity, variability and the genetic basis of innate immunity in the three-spined stickleback *Gasterosteus aculeatus*. Model parasite is the digenean trematode *Diplostomum pseudospathaceum*, which induces high fitness costs and is targeted primarily by the host's innate immune system. It is well suited for our purposes since it forms metacercariae in the fish's eye lens during its second larval phase, meaning that parasite load can be assessed non-invasively. We establish selection lines over at least two generations, which are challenged with homologous and heterologous parasite genotypes to assess evolutionary rates and specificity of host resistance. Trade-offs are investigated by infecting fish bred for enhanced resistance to *D. pseudospathaceum* with a different parasite, the tapeworm *Schistocephalus solidus*. We hypothesize that increased or lowered resistance resulting from the artificial selection regime is accompanied by heritable changes in innate immune gene expression. These changes are examined by analysing transcription patterns of all immunologically relevant genes.

Polymorphism of pathogenicity in species of *Verticillium* phytopathogens

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The genus *Verticillium* encompasses phytopathogenic species that cause vascular wilts of plants. It is a destructive disease with international consequences for many crops production, the better known species of *Verticillium* are, *V. dahliae* and *V. albo-atrum* cause a wilt disease. In my PhD research we will focus on *Verticillium dahliae* where the subject titled: Secondary metabolite production by *Verticillium dahliae* and their role on *Verticillium* wilt. The work aims to identification of new toxin produced by defoliating strain of *V. dahliae* which caused defoliating symptoms on infected susceptible olive species, then testing the possibility of natural transition of the genetic factors which are responsible of pathogenicity from defoliating strain to non-defoliating strain of *V. dahlia*.

Studies on the natural variation of the oil content of *Arabidopsis thaliana* seeds

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Increasing the oil content of oilseeds is an ongoing important task in plant breeding, considering the increased demand of plant oil for regenerative fuels and human consumption. The molecular step of storage oil synthesis are mostly known and many of the genes involved had been identified. However, most regulating genetic factors regarding the control and limitation of seed filling are still unknown. In this talk we will present our approach to identify new genes associated with seed oil content by making use of the natural variation of *Arabidopsis thaliana* and look into the element of epigenetic influence concerning this trait.

Conservation status of the marine otter (*Lontra felina*) in Peru - Field data and first genetic results

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The marine otter (*Lontra felina*), the smallest marine mammal of the world, is categorized as “Endangered” by the Red List of the IUCN, figures in Appendix I of CITES, Appendix I of CMS and is protected by Peruvian law. Its historical distribution, between 6°S and 56°S of the Southern Pacific coast, has been reduced in the last decades due to human factors such as habitat modification, pollution, bycatch and dynamite fishing. Although the species seems to adapt to habitat disturbances, the threats to the marine otter are constantly increasing. Little is known about social behavior and population evaluations show divergent results that range from 200 to 700 individuals at the Peruvian coast. For the effective conservation management of such an endangered species, population genetic studies are essential. Finally, the first genetic study about marine otters this study was conducted at the Peruvian coast between August and September of 2008. Otter scat was collected in 25 localities between 8°S and 18°S. DNA of 24 individuals was analyzed, showing a surprisingly high genetic variability (11 haplotypes, $h=0.86$ and $\pi=0.0117$). No evidence of substructuring, bottleneck or isolation by distance could be found. The adaptation to a naturally fragmented habitat could explain why, despite the reduction of numbers of individuals in the last decades, the species shows relatively high variability. According to predictions of population decrease and due to constant increase of threats we recommend to deepen the studies about the marine otter genetics, to monitor the populations periodically and to establish at least one protected area at the southern coast of Peru, where regions with high number of individuals are reported. These studies should not be limited by political barriers and should include the complete range of the species, comprising the countries of Peru, Chile and Argentina.

Phylogeography of the striped red mullet (*Mullus surmuletus* L.) inferred from microsatellite markers and mtDNA.

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The purpose of this work is to study the phylogeographic structure of the red striped mullet (*Mullus surmuletus*), using as molecular tools the microsatellite and the mitochondrial DNA. This species is of considerable economic importance in fisheries. It is naturally distributed from the north-west coasts of Africa and the Atlantic Ocean to the Black Sea and the Mediterranean. An enriched genomic library was developed for microsatellite-containing sequences (using an enrichment protocol) and primers sets were designed, optimized and applied on a *M. surmuletus* population as well as on four other species of the Mullidae family (*M. barbatus*, *Upeneus moluccensis*, *Pseudupeneus prayensis* and *Mulloidichthys martinicus*). Furthermore, twenty-four *M. surmuletus* populations, sampled from the North Sea to the Syrian coasts, were genotyped using a number of microsatellite loci. Moreover, certain individuals of each population were sequenced in order to obtain more information provided by mtDNA. Analyses strongly suggest the quasi-absence of genetic structure among the populations of *M. surmuletus* from the Atlantic Ocean and the Mediterranean Sea. The existence of three statistically differentiated genotype clusters ($p < 0.05$) into every *M. surmuletus* population was also revealed. Extensive gene flow and/or a recent population expansion could explain the observed population patterns.

Key words: Mullidae, *Mullus surmuletus*, microsatellite DNA, mitochondrial DNA, biogeography, genetic structure, cross-species amplification.

On the universality of weak selection

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Weak selection, which means a phenotype is slightly advantageous over another, is an important limiting case in evolutionary biology. Recently it has been introduced into evolutionary game theory. In evolutionary game dynamics, the probability to be imitated or to reproduce depends on the performance in a game. The influence of the game on the stochastic dynamics in finite populations is governed by the intensity of selection. In many models of both unstructured and structured populations, a key assumption allowing analytical calculations is weak selection, which means that all individuals perform approximately equally well. In the weak selection limit many different microscopic evolutionary models have the same or similar properties. How universal is weak selection for those microscopic evolutionary processes? We answer this mathematical question by investigating the fixation probability and the average fixation time not only up to linear, but also up to higher orders in selection intensity. We find universal higher order expansions, which allow a rescaling of the selection intensity. With this, we can identify specific models which violate (linear) weak selection results, such as the one-third rule of coordination games in finite but large populations.

Study on the genetic diversity of Citrus Tristeza Virus (CTV) populations in order to predict eventual disease outbreaks

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Citrus tristeza Closterovirus (CTV) has historically been one of the most devastating infectious diseases of citrus worldwide and currently still of major importance. The causal agent was identified as a monopartite, positive sense RNA genome of ca. 20Kb, the largest known of any plant virus, organized into 12 open reading frames (ORFs) and potentially coding for at least 19 proteins. Evolutionary phenotype expression of CTV is strictly governed by the structure of viral RNA populations frequently altered by subsequent recombination events. Additionally, vector transmission bottlenecks often contribute to sequence variant divergences which are sometimes accompanied by a variation of the pathogenic behaviour of the disease. Up to date, the virus characterization and virus-vector relationship have been a key factor in understanding the CTV genetic drift and epidemiology.

The overall goal of this research is the investigation of the genetic diversity of geographically distinct CTV isolates maintained in-planta in the Mediterranean Agronomic Institute of Bari (Italy), throughout multiple amplifications of similar sized DNA sequences, cloning and partial genome sequencing of the major capsid protein gene (p25).

Furthermore, aiming to evaluate eventual genetic variability by vector passage, some CTV isolates showing divergent genotype profiles were subjected to experimental transmission trials by a local aphid biotype of *Aphis gossypii* Glover. Viral templates targeting different genomic regions have been analysed by single strand conformation polymorphism (SSCP) and partial genome sequencing before and after transmission.

As a result, beside the presence of mixed infections, genome characterization analysis evidenced an abundance of mild isolates in the Mediterranean area. Furthermore, transmissibility trials indicated slight CTV genomic variations in the vector derived sub-isolates. Interestingly, this finding highlights the need for further investigations gathering the evolutionary behaviour of CTV populations in order to predict the disease dynamics and to prevent eventual outbreaks.