# Functional genomics in ecological model systems

**Professor Jim Marden** is a Professor of Biology at the Penn State University. He also co-directs the Huck Institute of Life Sciences and collaborates with researchers from Helsinki, Stockholm, Yale and Washington Universities. His recent work investigates how genetic traits affect ecological dynamics in natural systems.

n organism's success in any environment is a function of its fitness or suitability for that environment. This suitability is affected by a range of mechanical, evolutionary, physiological and ecological factors. The context of these factors changes over time, and over the life stages of the organisms. Professor Jim Marden and his team at Penn State University use genomics and physiological perspectives to understand 'functional genomics'. Their research focuses on physiological mechanisms, the functional effects of genetic variation, and how their associated traits affect the dynamics of wild populations and communities – in other words, how variation in traits affects fitness and competitive ability in natural habitats.

# METAPOPULATION DYNAMICS IN

Since 1992, biologists at the University of Helsinki have been studying a metapopulation (groups of same-species organisms separated by space) of Glanville fritillary butterflies (*Melitaea cinxia*), on the Aland Islands, Finland. In collaboration with Professor Marden, they discovered that this species is polymorphic (occurs in different forms) for traits that affect fitness in ways that are

dependent on habitat patch size, connectivity and population history. Discreet populations of the butterfly are found at around 400 sites in the area. On an annual basis, an average of 100 populations go extinct and 100 are newly formed. The research examines whether some genotypes, or genetic traits, are better adapted for dispersal and colonisation of new sites, while others are more suited to remain in an established patch. Which genes are responsible for these traits and how do they impact on the fitness of the butterfly populations?

# **ENERGETIC FOUNDERS**

The team found that daughters of the founders of new populations had a higher metabolic rate when compared to the daughters of the older, established populations. This suggests the 'founders' of the new populations had a higher peak performance and/or endurance. This was especially true in the patches of lowest connectivity – those sites more remote from others.

One reason for this is polymorphism of the gene which regulates Phosphoglucose Isomerase (PGI), the enzyme responsible for an early step in the breakdown of glucose to yield energy for activity. Two distinct

Long term ecological research systems are the equivalent of 'model organisms' for this new kind of research

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genotypes Pgi-A and Pgi-C were identified. Those organisms carrying the Pgi-C allele displayed higher flight metabolic rates and increased endurance, particularly at cooler temperatures. The frequency of the Pgi-C allele increases across the metapopulation where there is less habitat and it is more fragmented, suggesting that the organisms with the Pgi-C genotype are better at dispersing and forming new populations. This has been validated by field studies which show that female butterflies with a higher metabolic rate disperse farther than those with lower rates.

Polymorphism was also found in the metabolic enzyme succinate dehydrogenase (SDH), which regulates the hypoxia-inducible factor (HIF) pathway. Two variants of SDH show pronounced differences in flight performance which may be attributable to differences in tracheal network size, responses to low oxygen levels caused by flight, and mitochondrial health. The allele, or variant of SDH associated with a lower enzyme catalytic rate appears to cause development of a larger tracheal network in adult butterflies, with an associated higher metabolic output and slower rate of mitochondrial ageing. Possible tradeoffs with other traits, like larval growth rate, are presently under investigation.

Other work has shown that the butterflies from the new populations had a higher expression of genes involved in egg provision and in the maintenance of flight muscle proteins. Physiological studies confirmed that these new populations also have accelerated egg maturation. In all, this body of work has shown that ecological circumstances, in this case the patchiness of suitable habitat patches and the extinction/colonisation dynamics of local populations, maintains a balance of genetic variants and associated physiological traits.

# **FUNCTIONAL GENOMICS OF TROPICAL**

In Professor Marden's other ongoing research on tropical forests, a different collaborative team has been looking at the impact of microbial pathogens in the soil, and why this is more pronounced when the seedlings are growing next to conspecifics, or plants



of the same species. It is suggested that this prevents one or more species from becoming dominant in an ecosystem, due to the increased risk of growing near one's own kind and being susceptible to the same

This work aims to test the hypothesis that larger local populations have higher genetic diversity, reduced co-susceptibility and lower pathogen transmission. Conversely, smaller local populations may have reduced diversity, higher co-susceptibility, and therefore higher rates of disease transmission. If so, this may explain why ecological studies have repeatedly observed increased negative density dependence (NDD) in locally less common plant species. This would differ mechanistically from the classically held notion that common species suffer more from a build-up of species-specific pathogens and pests, but nonetheless provides a mechanism that prevents the domination of any one species in an ecosystem. As a species

becomes less common in the community, it is less likely to grow next to conspecifics and hence will not experience any negative consequence. Models have shown that such a pattern causes species to equilibrate at different abundances.

To begin to understand this at community level, roots of 75 seedlings of each of six species of tropical trees were examined for expression and allelic variation in all of their protein-coding genes. Genes that recognise and stimulate defence responses against pathogens (R genes) were of particular interest. This work found that polymorphism in R genes, which provides the ability to recognise genetically diverse pathogen strains, was significantly higher than other types of genes in the transcriptome, i.e., the total protein-coding genetic diversity of the plants. As predicted, the diversity of R-genes increases with local population size and is associated with reduced NDD. Gene expression data indicated that less common species were not deficient in their ability to mount defence responses when the sentinel function of R genes was bypassed by application of a plant defence hormone, but was reduced when the seedlings were challenged by live soil, a situation where recognition of diverse pathogens is essential.

Wild populations contain large amounts of non-neutral genetic variation that affects and is affected by ecological dynamics. One emergent message is that fitness is context dependant, varying not only with well-examined environmental variables like temperature and salinity, but also purely

biological features such as the age and

FITNESS IS CONTEXT DEPENDANT

size of local populations, which cannot be observed without extensive ecological study. These collaborative long-term ecological research programmes that incorporate functional genomics and physiology, replace the traditional approaches that attempt to understand organisms without reference to a detailed ecological context.

Professor Marden's ground-breaking approach, which considers genomics with ecological and evolutionary dynamics, produces a fine-grained knowledge of how functionally important variation works in



# Are the LTER models you have used with butterflies and forest trees transferable to other ecological systems?

Yes, but only with a lot of work. The butterfly metapopulation in Finland has been studied across a large spatial scale since 1992. The Forest Dynamics plot in Panama has identified, measured and mapped every stem >1cm diameter on a 50 hectare plot (>300,000 plants) every five years since 1985. A single researcher cannot readily do that on their own, but they can join forces with ecologists who have these kinds of study systems but lack a genomic and physiological perspective.

# As we know there is increased biodiversity in tropical forests, do the same population dynamics and NDD pressures apply to temperate and boreal

A relationship between NDD and local population size has been observed in temperature grasslands and forests, so it looks like the answer is "yes".

# If genetic diversity is increased within metapopulations, at a species level, what are the advantages for an organism to live in a continuous area with a reduced genetic variation?

Metapopulation refers to a structure in which the habitat is patchy and a species occurs only in patches. Distance between patches and limited flow of genetic variation, along with small numbers of individuals, reduces variation within each

Finally, the pathogens themselves answered

the question of "does this affect them?"; they

defences when in the more common species

expressed more of their own anti-oxidant

that have higher R gene diversity.

population (patch). So, it is correct that the metapopulation as a whole is more diverse than any of its constituent populations, but that does not mean it is more diverse than a structure in which more individuals are spread uniformly across the same area. The latter would contain more individuals, easier flow of genetic variation, and more total genetic variation.

# What applications does your work have for example in agriculture and disease prevention of crops?

We are using information about the diversity of R genes in wild tropical trees in a collaborative project that aims to improve disease resistance in cacao, the tropical tree from which we get chocolate. It turns out that R gene diversity in wild populations has been very little characterised, so there has never been an informed understanding of what is needed to breed crops that are resistant to a broad range of pathogens and strains within pathogen species. Note that crops are selectively bred for particular traits, which tends to remove genetic diversity across the genome, making crops highly vulnerable to pathogens. By identifying the R genes most likely to have alleles that are important, and using experimental approaches for verification, we think we can provide new ways to improve disease resistance in crops, particularly ones like tree crops with generation times too long to perform traditional breeding experiments.

# Detail

### **RESEARCH OBJECTIVES**

Professor Marden's research investigates development, physiology and how genetic and physiological variation affect the dynamics of ecological systems. His research foci include stoneflies, dragonflies, butterflies, and tropical

### **FUNDING**

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### COLLABORATORS

- Ilkka Hanski (University of Helsinki, deceased)
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- Claude dePamphilis (Penn State)
- Ruud Schilder (Penn State) • Liza Comita (Yale University)
- Scott Mangan (Washington University)
- Aleksandar Popadić (Wayne State
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    - Mark Guiltinan (Penn State)

A professor of Biology at Penn State University, Dr Marden combines genomics and physiological perspectives with long-term ecological data to understand the dynamics of some of the best studied natural systems, including fritillary butterflies in Europe and tropical trees in Panama.

## CONTACT

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# Fitness is context dependant: what works for an organism in one habitat may not work in another



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