

(1) The effect of migration on local adaptation in a coevolving host-parasite system.

Morgan et al (2005). *Nature*, 437, 253–256.

Antagonistic coevolution between hosts and parasites in spatially structured populations can result in local adaptation of parasites; that is, the greater infectivity of local parasites than foreign parasites on local hosts. Such parasite specialization on local hosts has implications for human health and agriculture. By contrast with classic single-species population-genetic models, theory indicates that parasite migration between subpopulations might increase parasite local adaptation, as long as migration does not completely homogenize populations. To test this hypothesis, we developed a system-specific mathematical model and then coevolved replicate populations of the bacterium *Pseudomonas fluorescens* and a parasitic bacteriophage with parasite only, with host only or with no migration. Here we show that patterns of local adaptation have considerable temporal and spatial variation and that, in the absence of migration, parasites tend to be locally maladapted. However, in accord with our model, parasite migration results in parasite local adaptation, but host migration alone has no significant effect. (154w)

The effect of differential migration rates on local adaptations in the DNA phage to its host *Pseudomonas fluorescens*

Marija

In antagonistic host-parasite coevolution, parasites are predicted to be locally adapted if they migrate more than their hosts, whereas hosts are predicted to be locally adapted if they migrate more than their parasites. In this study we examined the role of differential migration rates of hosts and parasites on parasite local adaptation with the use of bacterium *Pseudomonas fluorescens* as the host and associated lytic DNA phage SBW25φ2 as the parasite. We showed that parasite migration increased parasite local adaptation relative to unmigrated populations, but host migration did not decrease parasite adaptation. We also found differences in sympatric infectivity between migration treatments reflecting the introduction of genes that are beneficial for infecting a specific host population and the introduction of genes that are useful for phage infectivity across all populations. Our study showed that parasite migration can provide an evolutionary advantage to parasites in host-parasite coevolutionary race, and can therefore result in parasite local adaptation. Low rates of migration might increase within-population genetic variation of the migrated species, increasing evolutionary potential. (171w)

Parasite migration drives local adaptation of phage SBW02 to their host *Pseudomonas fluorescens*

Bea

Migration is one of the most important sources of genetic variation. However, how this process shapes the co-evolutionary dynamics of hosts and parasites interaction is not clear. Here, we address the impact of different migration rates of the bacteria *Pseudomonas fluorescens* and its phage, SBW02, in local adaptation. To do that, we performed in silico simulations and in vitro experiments in microcosms where independent bacteria and phage migration at different rates were performed for several generations. Parasite local adaptation was measure before the migration treatments and at six subsequent times as the performance of local parasites minus the average of foreign parasites. We have found that in both models phage migration increased phage local adaptation whereas bacteria migration seemed to have little effect. Besides, we have observed that effect of migration is asymmetric as host migration did not decrease parasite adaptation. On the contrary, no significant effect of migration was observed for host local adaptation. This is important as might reflect that as global interactions become stronger this not only will increase parasite transmission but also infectivity of parasites to their local host populations. (184w)

(2) Evolution of mutualism from parasitism in experimental virus populations.

Shapiro, et al (2018). *Evolution*, 72(3), 707–712.

While theory suggests conditions under which mutualism may evolve from parasitism, few studies have observed this transition empirically. Previously, we evolved *Escherichia coli* and the filamentous bacteriophage M13 in 96-well microplates, an environment in which the ancestral phage increased the growth rate and yield of the ancestral bacteria. In the majority of populations, mutualism was maintained or even enhanced between phages and coevolving bacteria; however, these same phages evolved traits that harmed the ancestral *E. coli* genotype. Here, we set out to determine if mutualism could evolve from this new parasitic interaction. To do so, we chose six evolved phage populations from the original experiment and used them to establish new infections of the ancestral bacteria. After 20 passages, mutualism evolved in almost all replicates, with the remainder growing commensally. Many phage populations also evolved to benefit both their local, evolving bacteria and the ancestral bacteria, though these phages were less beneficial to their co-occurring hosts than phages that harm the ancestral bacteria. These results demonstrate the rapid recovery of mutualism from parasitism, and we discuss how our findings relate to the evolution of phages that enhance the virulence of bacterial pathogens. (192w)

Evolution of mutualism are evolved from parasitism in the bacteria-phage interaction

Nenad

Mutualism depends on many biotic and abiotic factors. To understand the evolution of mutualism to parasitism a transition of both must be explored. Six evolved phage population from the original experiment had chosen in experiment to show reduction of the growth of an ancestral bacteria and *E. coli*. Four strain of *E. coli* have been used for insertion on the F plasmid for infection of *E. coli* by F-specific phage. The six virus populations were chosen for horizontal transmission and fully sequenced by using Illumina HiSeq platform. Results shows that the wild-type phage is beneficial to the ancestral *E. coli* whereas the founding phages that had evolved in the prior experiment are harmful to host. Phage populations of origin did not affect similar like their descendants affect the ancestral bacteria. The results of our study indicate that there can be a trade-off between benefiting the ancestral host and the beneficial of phage to help their hosts. This evolution can lead to phages capable of helping additional bacterial genotypes. (168)

Once enemies, now friends: Coresident phage M13 increases fitness of its host *E. coli*

Nik

Mutualistic relationships can evolve from essentially obligatory parasitic interactions, and vice versa. To explore these transitions across the continuum, we studied interactions between the filamentous phage M13 and its host, *E. coli*, which is known to exhibit reduced growth rates after the infection with this, also vertically transmitted, phage. Sequencing on Illumina platform to confirm initial variability, and standardised growth assays were performed in this host-parasite setting. We found that phages although having a harming effect at first actually increased fitness of its *E. coli* host in next generations. Demonstrating that parasitic phages can also bring selective advantage, we argue that better understanding of phage evolution and dynamics with its host is of essence for in-depth knowledge of understanding disease evolution and ecology. (123w)

(3) Running with the Red Queen: host-parasite coevolution selects for biparental sex.

Morran, et al (2011). *Science*, 333, 216–218.

Most organisms reproduce through outcrossing, even though it comes with substantial costs. The Red Queen hypothesis proposes that selection from coevolving pathogens facilitates the persistence of outcrossing despite these costs. We used experimental coevolution to test the Red Queen hypothesis and found that coevolution with a bacterial pathogen (*Serratia marcescens*) resulted in significantly more outcrossing in mixed mating experimental populations of the nematode *Caenorhabditis elegans*.

Furthermore, we found that coevolution with the pathogen rapidly drove obligately selfing populations to extinction, whereas outcrossing populations persisted through reciprocal coevolution. Thus, consistent with the Red Queen hypothesis, coevolving pathogens can select for biparental sex. (101w)

Antagonistic coevolution in novel environmental conditions under which outcrossing is favored and populations are interacting with a virulent pathogen

Vlasta

Outcrossing is the most prevalent model of reproduction among organisms. It can increase fitness and accelerate a population's rate of adaptation to novel conditions by permitting genetic exchange between diverse lineages, promoting genetic variation among offspring, and allowing beneficial alleles to be quickly assembled into the scale genome. As a model for the study was chosen examination on the nematode - *Caenorhabditis elegans* (composed of males and hermaphrodites) and its pathogenic bacteria - *Serratia marcescens*. The prior method to the experiment was mutagenized of the five independent replicate outcrossing populations of each mating type (obligate selfing, wildtype, obligate outcrossing) by exposing them to ethyl methane-sulfonate (EMS) to infuse novel genetic variation in each population and then the five replicate populations were passaged under three different parasite treatment: control, evolution and coevolution. Results showed that obligately selfing lineages were driven to extinction when driven to extinction when confronted with a coevolving parasite. The results also demonstrate macro and microevolutionary prediction of the Red Queen hypotheses, that sex can facilitate adaptation to novel environments, but the long-term maintenance of sex requires that the novelty does not wear off. (186w)

Host/parasite systems as models for testing the long-term maintenance of outcrossing

Maja Lazarević

Different mode of reproduction occurs among animals and plants, but the most dominant is outcrossing. Coevolutionary interactions between hosts and pathogens might generate ever-changing environmental conditions, and thus favour the long-term maintenance of outcrossing relative to self-fertilization or asexual reproduction. We chose to examine the *C. elegans*/*S. marcescens* system to generate antagonistic coevolution when a host population and a pathogen population are repeatedly passaged under selection together, thus permitting a direct test of the Red Queen hypothesis. The results demonstrate the ability of antagonistic coevolution to continually generate novel environment condition under which outcrossing is favoured and population persist when interacting with virulent pathogen. In summary, we found that sex can facilitate adaptation to novel environments, but the long-term maintenance of sex requires that the novelty does not wear off, which is consistent with the Red Queen hypothesis. (138w)

(4) Host-parasite “Red Queen” dynamics archived in pond sediment.

Decaestecker et al. (2007). *Nature*, 450, 870–873.

Antagonistic interactions between hosts and parasites are a key structuring force in natural populations, driving coevolution. However, direct empirical evidence of long-term host-parasite coevolution, in particular 'Red Queen' dynamics--in which antagonistic biotic interactions such as host-parasite interactions can lead to reciprocal evolutionary dynamics--is rare, and current data, although consistent with theories of antagonistic coevolution, do not reveal the temporal dynamics of the process. Dormant stages of both the water flea *Daphnia* and its microparasites are conserved in lake sediments, providing an archive of past gene pools. Here we use this fact to reconstruct rapid coevolutionary dynamics in a natural setting and show that the parasite rapidly adapts to its host over a period of only a few years. A coevolutionary model based on negative frequency-dependent selection, and designed to mimic essential aspects of our host-parasite system, corroborated these experimental results. In line with the idea of continuing host-parasite coevolution, temporal variation in parasite infectivity changed little over time. In contrast, from the moment the parasite was first found in the sediments, we observed a steady increase in virulence over time, associated with higher fitness of the parasite. (189w)

Red Queen dynamics in the interaction between *Daphnia magna* and its parasite *Pasteuria ramosa*.

Emanuele

Laminated lake sediments offer an opportunity to study evolutionary dynamics over many generations in freshwater invertebrates which can produce a dormant form. Those persistent forms can be cultivated again and used in empirical microevolutionary experiments. Such is the case of the cladoceran *Daphnia magna* and its bacterial parasite *Pasteuria ramosa*. Lake sediments sampled in the present study date back to 39 years in the past and provided many past dormant *D. magna* and *P. ramosa*. Such were cultivated in the laboratory to establish new clonal lineages. *D. magna* from each of the sampled layers was exposed to parasites coming either from lower, same or upper sediment layer. Therefore, exposing the cladoceran to past, contemporary or future parasites. *P. ramosa* infectivity was higher for contemporary host-parasite pairs, while past and future forms had lower infectivity. This shows that *P. ramosa* forms are better adapted to contemporary host forms but would lose such adaptation when infecting future *D. magna* lineages. This pattern is consistent with fast evolutionary changes between the parasite and its host. Therefore, a coevolution process mediated by fluctuating selection, which matches a Red Queen dynamic. (186w)

Long-term dynamics of *Daphnia*-parasite coevolution in a natural setting

Ayse

Host parasite interactions impose frequency dependent selections, however empirical data on temporal dynamics in natural settings are entirely lacking. It has so far remained impossible to document the long term dynamics of *Daphnia* parasite co-evolution in a natural setting. In this study we used two laminated lake sediments where *Daphnia magna* and its parasite *Pasteuria ramosa* produce propagule banks that provide unique possibility of reconstructing evolutionary dynamics. We carried out two cross infection experiments where we exposed *Daphnia* clones from different depths to parasites from different depths. Our results indicated fast evolutionary changes with parasites adapting to infect contemporary host genotypes. Simulations using realistic parameter settings showed parasite isolates from the near future were more infective and hosts were least infected when exposed to parasites from the recent past. In this historical reconstruction of host parasite interactions in a natural population, we provide empirical evidence for coevolution based on fluctuating selection. (151w)

(5) Local migration promotes competitive restraint in a host-pathogen “tragedy of the commons.”

Kerr et al (2006). *Nature*, 442, 75–78.

Fragmented populations possess an intriguing duplicity: even if subpopulations are reliably extinction-prone, asynchrony in local extinctions and recolonizations makes global persistence possible. Migration is a double-edged sword in such cases: too little migration prevents recolonization of extinct patches, whereas too much synchronizes subpopulations, raising the likelihood of global extinction. Both edges of this proverbial sword have been explored by manipulating the rate of migration within experimental populations. However, few experiments have examined how the evolutionary ecology of fragmented populations depends on the pattern of migration. Here, we show that the migration pattern affects both coexistence and evolution within a community of bacterial hosts (*Escherichia coli*) and viral pathogens (T4 coliphage) distributed across a large network of subpopulations. In particular, different patterns of migration select for distinct pathogen strategies, which we term 'rapacious' and 'prudent'. These strategies define a 'tragedy of the commons': rapacious phage displace prudent variants for shared host resources, but prudent phage are more productive when alone. We find that prudent phage dominate when migration is spatially restricted, while rapacious phage evolve under unrestricted migration. Thus, migration pattern alone can determine whether a de novo tragedy of the commons is resolved in favour of restraint. (197w)

Tragedy of the commons – evolutionary trajectory of host-pathogen interactions in *E. Coli* - Bacteriophage metapopulations

Dimitrija

Experimental evolution of interacting species in large metapopulations is comparatively unexploited. Using a model-host pathogen system we investigated how the pattern of migration within large metapopulations affects eco-evolutionary dynamics. *E. Coli* and viral pathogen phage T4 were embedded in two 96 well microplates. Population was perpetuated through transfer on 12h cycle and exposed to one of 3 migration regimes: restricted, unrestricted and well mixed. Additionally, two types of phage were analyzed: prudent and rapacious, to analyze the trade-off between productivity and competitive ability. Network topology may profoundly influence the evolutionary trajectory of host-pathogen interactions. We observed that population structure is critical to coexistence of hosts and their pathogens. Additionally, restricted migration tends to stabilize community dynamics but does not affect average density. In general, spatial restrictions in migration within a metapopulation can favour relatively cooperative use of the common resource, thus averting the tragedy of the commons. (148w)

Unraveling the migration effect in eco-evolution through predator-prey metapopulations modelling

Anna

The understanding of host-parasite evolution within the same population is critical for the understanding of epidemiology and evolution. The combined modeling and experimental approaches are required in order to discover the underlying dynamics, which have yet to be discovered. Here, we developed a host pathogen system in order to elucidate how the pattern of migration in large metapopulations affects eco-evolution. For that reason, we submitted bacteriophage T4 and *E. coli* into treatments with diverse abundance of each organism. Our findings strongly support the importance of structured populations for coexistence. Interestingly, we noticed lower phage density upon dilutions due to evolutionary pressure as the evolution of competitiveness leads to the lower productivity of phages. We extended our approach to 2 phage types, prudent and rapacious, and we concluded that they followed different strategies due to migration. Rapacious types showed lower overall productivity. Moreover, when dispersal between subpopulations was restricted, longstanding strategies were favoured, since highly connected social networks favor virulence. Overall, these findings suggest the key role of migration in large metapopulations and could as well be applied to other predator-prey interactions studies. (182w)

(6) Bacteria-phage antagonistic coevolution in soil.

Gomez & Buckling (2011). *Science*, 332, 106–109.

Bacteria and their viruses (phages) undergo rapid coevolution in test tubes, but the relevance to natural environments is unclear. By using a "mark-recapture" approach, we showed rapid coevolution of bacteria and phages in a soil community. Unlike coevolution in vitro, which is characterized by increases in infectivity and resistance through time (arms race dynamics), coevolution in soil resulted in hosts more resistant to their contemporary than past and future parasites (fluctuating selection dynamics). Fluctuating selection dynamics, which can potentially continue indefinitely, can be explained by fitness costs constraining the evolution of high levels of resistance in soil. These results suggest that rapid coevolution between bacteria and phage is likely to play a key role in structuring natural microbial communities. (119w)

Ecological and coevolutionary dynamics of *Pseudomonas fluorescens* and a naturally associated bacteriophage in soil

Tijana

Many ecological and evolutionary processes depend on the reciprocal evolution of host-parasite antagonistic coevolution. Coevolution of bacteria and viruses have been widely studied in the laboratory, but the information about bacteria and bacteriophage dynamics and consequence of coevolution is scarce. Bacteriophage only encounter hosts passively so it is unclear if they impose sufficient selection on bacteria to rapidly evolve. We tested ecological and evolutionary dynamics of *Pseudomonas fluorescens* and a naturally associated bacteriophage in soil in the presence and absence of natural community in space and time. We found that there is a difference between bacteria and phage coevolution in soil compared to laboratory. Bacteria and phage rapidly evolve in soil driven by fluctuating selection. Coevolutionary dynamics remains the same despite differences between the population dynamics of bacteria and phages in presence or absence of natural community. We haven't found differences in magnitude of parasite local adaptation in space and only small differences in adaptation in time. Coevolution altered ecological population dynamics and resulted in bacteria adapted to phages in time and phages adapted to bacteria in space. These results suggest that bacteria-phage dynamic coevolution is crucial in explaining the structure, population dynamics and the function of natural microbial communities. (200w)

A Comparative Method to Determine Bacteria-Phage Coevolution Driven by Fluctuating Selection

Evrin

The antagonistic coevolution between host and parasite is important for ecological and evolutionary processes like population dynamics, extinction risk, speciation, diversity and evolution of sex and mutation rates. Especially the antagonistic coevolution of bacteria and phages has a broad impact on ecosystem functioning and therapeutic use. Therefore we compare the evolution of a soil bacteria clone, *Pseudomonas fluorescens* SBW25 and its bacteriophage clone SBW25ø2 in soil microcosms with the presence and absence of natural community. We look at the mean densities of the populations first, then we measured the infectivity of bacteria and phage at different time points (day-0, day-24 and day-48). Lastly, we tested the hypothesis that resistance is more costly in soil by comparing phage-resistant and -sensitive clones of bacteria in both broth and soil environments. We showed that bacteria and phage rapidly coevolve in soil with very similar dynamics in the presence and absence of the natural community. We also found that bacteria are most resistant at contemporary time while phages are most ineffective. Our results suggest that rapid bacteria-phage coevolution are likely to be crucial in explaining the structure, population dynamics and ultimately the function of natural microbial communities. (193w)

(7) Effects of shortened host life span on the evolution of parasite life history and virulence in a microbial host-parasite system.

Nidelet et al, 2009 *BMC Evolutionary Biology*, 9, 65

BACKGROUND: Ecological factors play an important role in the evolution of parasite exploitation strategies. A common prediction is that, as shorter host life span reduces future opportunities of transmission, parasites compensate with an evolutionary shift towards earlier transmission. They may grow more rapidly within the host, have a shorter latency time and, consequently, be more virulent. Thus, increased extrinsic (i.e., not caused by the parasite) host mortality leads to the evolution of more virulent parasites. To test these predictions, we performed a serial transfer experiment, using the protozoan *Paramecium caudatum* and its bacterial parasite *Holospira undulata*. We simulated variation in host life span by killing hosts after 11 (early killing) or 14 (late killing) days post inoculation; after killing, parasite transmission stages were collected and used for a new infection cycle. **RESULTS:** After 13 cycles (approximately 300 generations), parasites from the early-killing treatment were less infectious, but had shorter latency time and higher virulence than those from the late-killing treatment. Overall, shorter latency time was associated with higher parasite loads and thus presumably with more rapid within-host replication. **CONCLUSION:** The analysis of the means of the two treatments is thus consistent with theory, and suggests that evolution is constrained by trade-offs between virulence, transmission and within-host growth. In contrast, we found little evidence for such trade-offs across parasite selection lines within treatments; thus, to some extent, these traits may evolve independently. This study illustrates how environmental variation (experienced by the host) can lead to the evolution of distinct parasite strategies. (250w)

Shortened host life span cause adaptive shift in parasite latency time and increase parasite virulence

Jelisaveta

Increased extrinsic host mortality is thought to select greater parasite growth rate, followed by earlier transmission, and stronger virulence. Previous studies showed that shortening the life span of a host stimulate the virus response, but not always as predicted by the theory. Here we investigate the effect of early versus late killing of *Paramecium caudatum*, a host of bacterial parasite *Holospira undulata* on different aspects of parasite life history. We manipulated host life span in two treatments; early-killing and late killing, by killing hosts after 11 or 14 days upon infection, respectively. Subsequently, we compared parasites for their infectivity, latency time, parasite load and virulence. Consistent with the predictions, parasites from the early- killing treatment had a shorter latency time and were more virulent compared to a late- killing host. Shorter latency time was likely a result of faster within-host replication. Our study provides insights into how variation in environmental conditions affecting host life- history can influence the evolution of parasite life history. (163w)

Increase in virulence of a bacteria caused by a shortened host life span

Jelena

A shorter host life may cause an increased virulence of parasites by making them grow and reproduce faster in order to compensate the loss of future transmission. We tested this hypothesis by experimentally manipulating a host life span by having two treatments - early-killing (after 11 days) and late-killing (after 14 days) and used the parasites from these two treatments for future infections. After 13 infection cycles, we compared the parasites from the two treatments for their infectivity, latency time, parasite load and virulence. Early-kill parasites had a shorter latency time, and they went into the infective phase faster which resulted in a 10% larger parasite load. Host lines infected with early-kill parasites had significantly lower densities, mainly due to a 32% increase in population mortality. This study confirms the initial presumption that a change in host life history can impact the evolution of parasite life history, and in this case a shorter host life span resulted in a faster development and increased virulence of parasites. (166w)

(8) Effects of predation on real-time host-parasite coevolutionary dynamics.

Friman & Buckling (2013). *Ecology Letters*, 16, 39–46.

The impact of community complexity on pairwise coevolutionary dynamics is theoretically dependent on the extent to which species evolve generalised or specialised adaptations to the multiple species they interact with. Here, we show that the bacteria *Pseudomonas fluorescens* diversifies into defence specialists, when coevolved simultaneously with a virus and a predatory protist, as a result of fitness trade-offs between defences against the two enemies. Strong bacteria-virus pairwise coevolution persisted, despite strong protist-imposed selection. However, the arms race dynamic (escalation of host resistance and parasite infectivity ranges) associated with bacteria-virus coevolution broke down to a greater extent in the presence of the protist, presumably through the elevated genetic and demographic costs of increased bacteria resistance ranges. These findings suggest that strong pairwise coevolution can persist even in complex communities, when conflicting selection leads to evolutionary diversification of different defence strategies. (139w)

Community complexity affects host-parasite coevolutionary dynamics and defence strategies in *Pseudomonas fluorescens*

Branka

Coevolution plays an important role in structuring the natural communities, which further alter the reciprocal selection pressures and coevolution dynamics. We studied how selection by predators (*Tetrahymena thermophila*) and parasites (bacteriophage) affect the evolutionary diversification of prey (*Pseudomonas fluorescens*) defence strategies and how pairwise coevolution between host and parasite is changed in the predator's presence. *P. fluorescens* in medium served as a control treatment, and experimental groups were inoculated with phage, with protists or with combination of both. Bacterial and phage population densities and number of protists were determined after 28 days. Bacterial resistance and phage infectivity, bacterial defence against protists and bacterial growth measurements were analysed with General Linear Mixed Models. Results show bacterial defence against both enemies evolved to be greater in single-enemy communities than in the presence of both. Concurrent selection by two enemies led to divergence of specialised defence against them. Coevolution of bacteria and phage showed short-term arms race dynamics (increased bacterial resistance and phage infectivity throughout time), but it was weakened in the presence of the protist and shifted towards fluctuating selection dynamic. Demography and trade-offs are the two important factors contributing to this shift. Defence against one enemy constrained the defence against the other. (200w)

Investigation of coevolutionary dynamics of complex communities in real-time

Ayshin

Coevolution, the case where two or more species reciprocally affect each other's evolution could influence natural communities' structure and function. Community structure is one of the main factors that affects the pressure of selection among different species. Presence of multiple enemies in a community could result in evolution of either a specialist or generalist defense strategy. We investigated the changes in pairwise coevolutionary dynamics between a host (*P. fluorescens*) and a parasite (bacteriophage $\Phi 2$) after addition of a predator (*T. thermophila*) in real-time which has not been investigated before. It turns out that presence of a predator makes *P. fluorescens* to develop the specialist defense strategy due to the variations in selection pressure through time and space. Coevolution of *P. fluorescens* and bacteriophage $\Phi 2$ continued although the arms race dynamics between them weakened because of the *T. thermophila*. This study shows the importance of real-time coevolutionary experiments in explaining the observed discrepancy of phenotypic traits of species that coevolve in the same geographical area. (159w)

(9) Long-term selection experiment produces breakdown of horizontal transmissibility in parasite with mixed transmission mode.

Dusi et al (2015). *Evolution*, 69, 1069–1076.

Evolutionary transitions from parasitism towards beneficial or mutualistic associations may encompass a change from horizontal transmission to (strict) vertical transmission. Parasites with both vertical and horizontal transmission are amenable to study factors driving such transitions. In a long-term experiment, microcosm populations of the protozoan *Paramecium caudatum* and its bacterial parasite *Holospora undulata* were exposed to three growth treatments, manipulating vertical transmission opportunities over ca. 800 host generations. In inoculation tests, horizontal transmission propagules produced by parasites from a 'high-growth' treatment, with elevated host division rates increasing levels of parasite vertical transmission, showed a near-complete loss of infectivity. A similar reduction was observed for parasites from a treatment alternating between high growth and low growth (i.e., low levels of population turn-over). Parasites from a low-growth treatment had the highest infectivity on all host genotypes tested. Our results complement previous findings of reduced investment in horizontal transmission and increased vertical transmissibility of high-growth parasites. We explain the loss of horizontal transmissibility by epidemiological feedbacks and resistance evolution, reducing the frequency of susceptible hosts in the population and thereby decreasing the selective advantage of horizontal transmission. This illustrates how environmental conditions may push parasites with a mixed transmission mode towards becoming vertically transmitted non-virulent symbionts. (202w)

Transmission mode shift of *Holospora undulata* is coupled with its transition from parasitism to nonvirulence

Nemanja

The transmission mode of parasites, implicated in their spread and virulence, represents one of the key factors shaping their interactions with hosts. Due to their different virulence optima, horizontal and vertical transmission are suspected to drive the evolution of parasitism and mutualism, respectively. By manipulating growth conditions of *Holospora undulata*, a parasite of *Paramecium caudatum* with a mixed transmission mode, we tested the evolution of its horizontal transmissibility by exposing it to selection regimes with different relative importance of vertical to total transmission. Additionally, we evaluated the evolution of host response by comparing the resistance of paramecia which were exposed to *H. undulata* and those who were not. Our findings showed that after roughly 800 host generations of selection, there was a near-complete breakdown of horizontal transmissibility of *H. undulata* in conditions favoring frequent vertical transmissibility and the evolution of nonvirulence. We also confirmed the parasite-mediated selection for increased resistance, with paramecia from infected selection lines being relatively more resistant to infection. These findings are discussed in light of evolutionary and epidemiologic feedbacks acting upon host-parasite interactions and the importance of the changes in transmission mode of parasite in the macroevolutionary transitions between parasitism and mutualism. (196w)

Evolution of *Holospora undulata* horizontal transmissibility loss in populations of *Paramecium caudatum*

Ivana

Many parasites are capable of horizontal and vertical transmission, depending on trade-offs between two modes and ecological and epidemiological factors. Horizontal transmission should increase at high host population densities, while vertical is expected in hosts with higher fecundity. We manipulated growth conditions to change relative importance of vertical to horizontal transmission of the bacterial parasite *Holospora undulata* in populations of *Paramecium caudatum*. We imposed three different treatments (high-growth, low-growth and alternating) on infected and uninfected selection lines, and horizontal transmissibility was assessed using logistic regression models. Also we compared infectivity of high-growth and low-growth parasites on different host genotypes. Parasite *H. undulata* almost completely lost horizontal transmissibility after 800 host generations in the high-growth treatment. Alternating parasites followed the same trajectory as high-growth parasites (but at a slower rate), while infection prevalence was lower in low-growth treatment. The infectivity breakdown occurred independently across the multiple selection lines, suggesting that it was the result of selection rather than random loss of function. Environmental conditions had strong impact on evolution of the bacterial parasite transmission pathway, pushing it toward becoming a vertically transmitted nonvirulent symbiont. (183w)

(10) Antagonistic coevolution accelerates molecular evolution.

Paterson, S. et al. (2010). *Nature*, 464, 275–278.

The Red Queen hypothesis proposes that coevolution of interacting species (such as hosts and parasites) should drive molecular evolution through continual natural selection for adaptation and counter-adaptation. Although the divergence observed at some host-resistance and parasite-infectivity genes is consistent with this, the long time periods typically required to study coevolution have so far prevented any direct empirical test. Here we show, using experimental populations of the bacterium *Pseudomonas fluorescens* SBW25 and its viral parasite, phage Phi2 (refs 10, 11), that the rate of molecular evolution in the phage was far higher when both bacterium and phage coevolved with each other than when phage evolved against a constant host genotype. Coevolution also resulted in far greater genetic divergence between replicate populations, which was correlated with the range of hosts that coevolved phage were able to infect. Consistent with this, the most rapidly evolving phage genes under coevolution were those involved in host infection. These results demonstrate, at both the genomic and phenotypic level, that antagonistic coevolution is a cause of rapid and divergent evolution, and is likely to be a major driver of evolutionary change within species. (186w)

Antagonistic coevolution as a driver of molecular evolution

Maria

It has been proposed that when adaptation increases the fitness of one species, this necessarily causes a decline in fitness of other ecologically interacting species. These dynamics are rather important in the antagonistic co-evolution of hosts and virulent parasites. We have previously demonstrated that bacterium *Pseudomonas fluorescens* and its viral parasite, phage $\Phi 2$, undergo a persistent coevolutionary ‘arms race’ and now we are interested in studying the link between this rapid phenotypic evolution and the underlying pattern of molecular evolution. Here, we report a higher observed rate of evolution in the phages that were coevolving with the host against those that were evolving against a constant bacterium genotype. Coevolved phage populations also demonstrate a higher genetic divergence from their ancestor as that of the evolved populations. We argue that our findings suggest that antagonistic coevolution causes rapid evolutionary changes within species while creating sufficient between-population genetic divergence. (148w)

(11) High parasite diversity accelerates host adaptation and diversification.

Betts et al (2018). *Science*, 360, 907–911.

Many antagonistic interactions between hosts and their parasites result in coevolution. Although coevolution can drive diversity and specificity within species, it is not known whether coevolutionary dynamics differ among functionally similar species. We present evidence of coevolution within simple communities of *Pseudomonas aeruginosa* PAO1 and a panel of bacteriophages. Pathogen identity affected coevolutionary dynamics. For five of six phages tested, time-shift assays revealed temporal peaks in bacterial resistance and phage infectivity, consistent with frequency-dependent selection (Red Queen dynamics). Two of the six phages also imposed additional directional selection, resulting in strongly increased resistance ranges over the entire length of the experiment (ca. 60 generations). Cross-resistance to these two phages was very high, independent of the coevolutionary history of the bacteria. We suggest that coevolutionary dynamics are associated with the nature of the receptor used by the phage for infection. Our results shed light on the coevolutionary process in simple communities and have practical application in the control of bacterial pathogens through the evolutionary training of phages, increasing their virulence and efficacy as therapeutics or disinfectants. (175w)

Host-parasite coevolution: Influence of diverse parasite communities on evolution of the host

Katarina

Host-parasite coevolution operates within a network of species interaction. The parasite has been shown to play a key role in evolutionary diversification within and among host populations and coevolution with highly diverse parasite communities could ultimately drive faster host evolution and increased divergence among host population. To test this hypothesis, we exposed *Pseudomonas aeruginosa* to communities of one to five lytic viral bacteriophages and sequenced entire host and parasite populations longitudinally at multiple time points. We found 474 nonsynonymous and 75 synonymous polymorphisms across 173 gene and 133 intergenic mutations. We observed parallel evolution in genes that are known phage receptors. Parasite diversity does not affect genetic diversity within the host population, but have a profound effect on allopatric diversification among the host population (ANOSIM, $R=0.11$, $P < 0.01$). Our study reveals that parasite communities can form hotbeds of rapid antagonistic coevolution. (142w)

Coevolutionary interactions of Host with multiple parasites

Pavle

Host parasite coevolution is traditionally investigated in a framework where a single parasite species infects one host species, however in nature hosts are often under attack by multiple parasite species and parasites must compete for hosts. We exposed host bacterium *Pseudomonas aeruginosa* to communities of one to five lytic viral parasites (bacteriophages). Bacterial resistance increased with parasite diversity and parasite infectivity decreased, suggesting that parasite communities impose stronger selection on their host than single parasites alone. We observe parallel evolution common in all treatments in genes that are known phage receptors including genes for LPS, type IV pili and the Ton-B-dependent receptor. On average host alleles with higher frequencies tended to decline in frequency over time in the low and medium parasite diversity treatment, consistent with negative frequency-dependent selection, which is in accordance with the Red Queen dynamics. Strong directional selection was more common as parasite diversity increased (arms race) such that the number of fixed alleles in the high diversity treatment was more than two times that observed under medium diversity, OF the 29 alleles approaching fixation, 23 were in genes relating to LPS biosynthesis, which may provide general phage resistance mechanism. (195w)

(12) Antagonistic competition moderates virulence in *Bacillus thuringiensis*.

Garbutt et al (2011). *Ecology Letters* doi:10.1111/j.1461-0248.2011.01638.x

Classical models of the evolution of virulence predict that multiple infections should select for elevated virulence, if increased competitiveness arises from faster growth. However, diverse modes of parasite competition (resource-based, antagonism, immunity manipulation) can lead to adaptations with different implications for virulence. Using an experimental evolution approach we investigated the hypothesis that selection in mixed-strain infections will lead to increased antagonism that trades off against investment in virulence. Selection in mixed infections led to improved suppression of competitors in the bacterial insect pathogen *Bacillus thuringiensis*. Increased antagonism was associated with decreased virulence in three out of four selected lines. Moreover, mixed infections were less virulent than single-strain infections, and between-strain competition tended to decrease pathogen growth *in vivo* and *in vitro*. Spiteful interactions among these bacteria may be favoured because of the high metabolic costs of virulence factors and the high risk of mixed infections. (145w)

Influence of mixed-infections on the evolution of virulence

Milica

The evolution of increased competitive ability was associated with reduced virulence. Single strain-infections were more virulent strain than mixed-strain infections which tended to reduce pathogen density. In these competition experiment, a pathogenic and a non-pathogenic strain (*Bacteria thuringienisis*) for *Platella xylostela* were used. Several independent experiments showed that single-strain infections were more virulent and tended to increase bacterial growth. Evolutionary modeling predicts that co-operators will evolve lower levels of investments in traits for which is reduced relatedness. The presence of competitor reduced total bacterial productivity. Increased antagonism was associated with decreased virulence in three out of four selected lines. The presence of competitor affected bacterial growth and is it or not increased competitive ability which is associated with fitness cost. The effects of relatedness at toxin loci may have had small influence on the results of this study it is likely that investment in antagonism and public goods are linked in many bacterial pathogens. (154w)

Experimental evolution of pathogenicity traits of *Bacillus thuringiensis* in single-strain and mixed-strain infection conditions

Lea

Parasite competition within host during mixed infection can have broad effect on pathogenic traits. This competition is often mediated via investment in spite - by secreting antagonists, like antibiotics and bacteriocins, which lower the fitness of distantly related neighbours. These investments are expected to be metabolically costly, and as such should lead to decreased virulence in single infections and changes in other parasitic characteristics. Our study uses experimental evolution to investigate effects of selection in mixed and single-strained infections on pathogen virulence and productivity as well as investment in spite. We used two strains of important biological control agent, *Bacillus thuringiensis*, as pathogens, of which one had a role of a competitor, and selected them for improved reproduction in both regimes. We tested pathogens from different selection regimes for bacteriocines, and assessed their growth, fitness and virulence via *in vitro* competition and pathogen virulence assays. We found that *Bacillus* strains do act antagonistically through bacteriocines. We showed that pathogens selected in mixed-strain infections are better at suppressing competitors, but have reduced densities and virulence compared to those selected in single-strain regime. These results suggest that investing in spite is costly, thus decreasing amount of energy needed for growth and virulence. (200w)