domestic rabbits in China. The recombination signal is only observed in Nigeria camel-HKU23, suggesting a regional varied evolutionary history of camel-HKU23. Our findings extended the knowledge of the evolutionary relationships among Group 2a CoVs. Further surveillance in other African camels will be important to elucidate the evolution of camel-HKU23.

**Molecular systematics of sturgeon nucleocytoplasmic large DNA viruses**

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Namao virus (NV) is a sturgeon nucleocytoplasmic large DNA virus (nSNCLDV) that can cause a lethal disease of the integumentary system in lake sturgeon Acipeinerus fulvescens. As a group, the nSNCLDV have not been assigned to any currently recognized sncDNA viruses. In this study, a dataset of NV DNA sequences was generated and assembled as two non-overlapping contigs of 306 and 448 base pairs (bp) and then used to conduct a comprehensive systematic analysis using Bayesian phylogenetic inference for NV, other nSNCLDV, and representative members of six families of dsDNA virus superfamily. The phylogeny of NV was reconstructed using protein homologues encoded by nine nucleocytoplasmic virus orthologous genes (NCVOs): NCVOG00224—mcp, NCVOG00388—DNA polymerase B elongation subunit, NCVOG00726—V A1R-type helicase, NCVOG02439—V A32-type ATPase, NCVOG02626—AL2 VLT3-like transcription factor, NCVOG02719—RNA polymerase II subunit II, NCVOG02749—RNA polymerase II subunit I, NCVOG02746—ribonucleotide reductase small subunit, and NCVOG1117—mRNA capping enzyme. The accuracy of our phylogenetic method was evaluated using a combination of Bayesian statistical analysis and congruence analysis. Stable tree topologies were obtained with datasets differing in target molecule sequence length, and taxa. Congruent topologies were obtained in phylogenies constructed using individual protein datasets and when four proteins were used in a concatenated approach. The major capsid protein phylogeny indicated that ten representative nSNCLDV form a monophyletic group comprised of four lineages within a polyphyletic Mimi-Phycodnaviridae group of taxa. Overall, the analyses revealed that Namao virus is a member of the Mimi-Phycodnaviridae family with strong and consistent support for a clade containing NV and CroV as sister taxa.

**Visualization of recombination in deformed wing virus infecting bees**

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Honey bees suffer increasing colony mortality worldwide, partially caused by the spread of viral pathogens. Among these pathogens, deformed wing virus (dWV) is one of the major, widespread viruses of honey bees resulting in wing deformities and weakening colonies. dWV can be found in honey bees, bumble bees, and other wild bees as three major genotypes named dWV-A, -B (also named Varroa destructor virus 1), and -C. Various recombinants of dWV-A and -B have been previously found in honey bees, some of which have been suggested to have higher virulence over non-recombinant, parental virus. In most of these cases, recombinants were only shown as consensus sequences from previous assemblies and alignments and may not reflect the biological reality of the recombinant present within a host bee. It is therefore important to build a method of recombinant detection and quantification within mixed infections in single-host individuals, including both parental and various recombinant genomes, so as to evaluate the relevance of recombinants for viral genome evolution and the impact on hosts. Here, we propose to visualize and quantify these recombinants using next-generation sequencing data to better understand how these genomes evolve within bees. Our method will be performed directly from raw sequence reads from various datasets (including field and lab experiments as well as screening of public data) in order to obtain an overview of dWV recombinant in various in vivo and in vitro conditions. Recombination of viral genomes is a key point for virus evolution. The detection and quantification of recombination will facilitate analysis of the determinants of recombination and help in understanding the routes by which new viral variants emerge. The emergence of new (more virulent) recombinant viruses can result from acquisition of new capabilities, such as escape from host immunity or increased transmission rates. Recombination can also lead to adaptation to new environments and new hosts by a change in cell tropism, allowing cross-species transmission, which may be particularly relevant for bumble bees and wild bees infected by honey bee-derived dWV.

**An evolutionary framework to guide the hunt for human dsDNA viruses**

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It is becoming increasingly obvious that we only know a fraction of the human virome. The development of next-generation sequencing (NGS) technologies has dramatically increased our ability to hunt viruses. Yet, the small genomes and low copy numbers characteristic of most viruses make undirected (shotgun) hunts a relatively inefficient strategy. Here, we propose to step up the rate of double-stranded DNA (dsDNA) virus discovery by combining NGS with evolutionary thinking. dsDNA viruses are thought to have co-diverged with their hosts. As this applies to the homineline lineages (gorillas, humans, chimpanzees, and bonobos), it is theoretically possible to estimate the phylegenetic position of cryptic human viruses by identifying co-divergent viral lineages infecting non-human hominines. Where these lineages do not comprise a human-infecting counterpart, a yet-unknown human virus may be hiding. The first phase of this project will consist in the high-throughput characterization of dsDNA viruses (herpesviruses, papillomaviruses, and polyomaviruses) infecting wild gorillas, chimpanzees, and bonobos. For this, we will use an exhaustive collection of fecal samples (in terms of hominine species/sub-species diversity) and apply a discovery strategy combining in-solution capture and NGS. This strategy has been developed in the ancient DNA field but has a very broad applicability; it will constitute a nice addendum to the institute technical protocol. Thanks to the massive amount of information collected, we will be able to reconstruct the evolutionary history of many dsDNA virus lineages and to identify those where a human virus would be expected but is still unknown. This will pave the way to the second phase of the project which will consist in a pre-oriented dsDNA virus human hunt based on the use of specific PCR systems implemented in multiplex. We expect that this project will generate an unprecedented amount of data on the processes at play along dsDNA virus evolution (co-divergence versus cross-species transmission), help determine the directionality, frequency, and timing of cross-species transmission events between hominines and unveil the existence of yet-to-be-discovered human viruses.

**Epidemic dynamics of ancient disease outbreaks**

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Bayesian phylogenetic analysis allows for the estimation of the time to the most recent common ancestor (tMRCA) of sequences sampled at different times, as long as they prove to be ‘measurably evolving’, which means that the time between sampling dates was long enough to allow the appearance of a measurable amount of genetic changes. This ‘temporal signal’ can be tested with the software TempEst (Rambaut et al. 2016), which generates a regression of the root-to-tip genetic distance on sampling times and finds the best-fitting root that produces the lowest residual sum of squares. For the case of pathogen single nucleotide
A strategy to studying zoonotic infectious diseases

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An increasing number of zoonotic viruses have been detected in animals, especially in poultry species. Understanding the diversity of zoonotic infections and the local behavior helps to characterize the pathogen diversity in human and animal species. The risk of pathogen spill-over from animals to humans is considerable, along with other countries in Southeast Asia, as a hotspot for zoonotic viruses. In Vietnam, domestic animals are typically farmed in close proximity to humans, which may increase the risk of transmission of zoonotic pathogens. Our previous studies found the presence of some zoonotic viruses (e.g. rotavirus group A, hepatitis E virus) in domestic pigs. However, the risk of pathogenic transmission from domestic animals to humans has not been determined. Detailed genomic sequence data may help to track the origin and evolution of zoonotic viruses.

Revealing the evolution of virulence in RNA viruses

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A combination of high rates of mutation and replication, coupled with strong natural selection, ensures that RNA viruses experience rapid genotypic and phenotypic evolution. Such a ‘fast-forward’ evolution enables viruses to rapidly adapt to new host species, evade host immune responses, and to develop resistance to antiviral drugs. Similarly, rapid evolution allows viruses to attain new levels of virulence, defined as the ability to cause severe disease in hosts. We hypothesize that distinct viral groups share genetic determinants that modulate virulence that have been acquired through convergent evolution. Thus, common patterns reflecting changing virulence-related specific viral groups could be detected. The main goals for this project are (1) to understand how genetic and phenotypic diversity can be generated among different viral groups by analyzing the variation patterns and determining the selective forces behind them (impact in viral fitness) and (2) to understand how fixed mutations can modulate virulence within different viral groups by performing comparison of strains with differing virulence within a longitudinal timescale. The subject of the study is key emerging and re-emerging virus families of medical importance. Such groups include: Coronaviridae (severe acute respiratory syndrome and Middle East respiratory syndrome-associated coronaviruses), Picornaviridae (Hepatitis A virus), Flaviviridae (Yellow fever, West Nile, Hepatitis C, Dengue, and Zika viruses), Togaviridae (Rubella and Chikungunya virus), Bornaviridae (Borna-disease virus), Filoviridae (Ebola and Marburg viruses), Paramyxoviridae (Measles, Nipah, and Hendra viruses), Retroviridae (Lentiviruses), and Influenza viruses. Understanding how fixed mutations can modulate virulence within different viral groups is crucial for understanding diversity patterns among clades, regions, and hosts.

A major likelihood-based approach gives problematic estimates of diversification dynamics and rates

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The diversity of life is shaped by rates of speciation and extinction, and so estimating these rates correctly is crucial for understanding diversity patterns among clades, regions, and habitats. In 2011, Morlon and collaborators developed a promising likelihood-based approach to estimate speciation and extinction and to infer the model describing how these rates change over time based on AICc. This approach is now implemented in A R package (RPAANDA). Here, we test the accuracy of this approach under simulated conditions, to evaluate its ability to correctly estimate rates of speciation, extinction, and diversification (speciation—extinction) and to choose the correct underlying model of diversification (e.g. constant or changing rates of speciation and extinction over time). We found that this likelihood-based approach frequently picked the incorrect model. For example, with changing speciation rates over time, the correct model was chosen in only ~10 per cent of replicates. There were significant relationships between true and estimated speciation rates using this approach, but relationships were weak when speciation rates were constant within clades.