Relationships were consistently weak between true and estimated rates of extinction and of diversification. Overall, we suggest that results from this approach should be interpreted with considerable caution.

A63 Quantifying the dynamics of evolutionary rates through time

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The availability of evolutionary rate estimates in recent years led to the observation that they may depend on the time-scale on which they are measured. Specifically, RNA virus evolutionary rates are frequently estimated to be low towards the past and high towards the present. This time-dependent rate phenomenon (TDRP) has important implications for evolutionary studies as it could severely bias divergence time estimates. While recent studies are providing insights into the relationship between viral evolutionary rate and time, formal probabilistic models to draw inference under TDRP scenarios remain lacking. Here, we adopt epoch-modelling to develop a Bayesian model of discrete rate changes through time in an unknown evolutionary history and combine this with a log-linear parameterization of rates as a function of times in the past. We provide an implementation for nucleotide substitution rates as well as for nonsynonymous rates change in a codon substitution model. Using a foamy virus dataset for which internal node calibrations can be applied based on host-virus co-divergence, we estimate a significant decline in evolutionary rates as a function of time into the past for nucleotide substitutions as well as for non-synonymous substitutions in a codon model. We also estimate a deep evolutionary history for primate Lentiviruses by combining an HIV-1 group M node calibration and a biogeographic calibration for viruses in drill monkeys in the TDRP model. Our analyses lead to the conclusion that studies of evolutionary timescales require a reconsideration of substitution rates, in either codon and nucleotide substitution model, as a dynamic feature of molecular evolution.

A64 Viral sequence classification using deep learning algorithms

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Sewage samples have a high potential benefit for surveillance of circulating pathogens because they are easy to obtain and reflect population-wide circulation of pathogens. These type of samples typically contain a great diversity of viruses. Therefore, one of the main challenges of metagenomic sequencing of sewage for surveillance is sequence annotation and interpretation. Especially for high-threat viruses, false positive signals can trigger unnecessary alerts, but true positives should not be missed. Annotation thus requires high sensitivity and specificity. To better interpret annotated reads for high-threat viruses, we attempt to determine how classifiable they are in a background of reads of closely related low-threat viruses. As an example, we attempted to distinguish poliovirus reads, a virus of high public health importance, from other enterovirus reads. A sequence-based deep learning algorithm was used to classify reads as either polio or non-polio enterovirus. Short reads were generated from 500 polio and 2,000 non-polio enterovirus genomes as a training set. By training the algorithm on this dataset we try to determine, on a single read level, which short reads can reliably be labeled as poliovirus and which cannot. After training the deep learning algorithm on the generated reads we were able to calculate the probability with which a read can be assigned to a poliovirus genome or a non-poliovirus genome. We show that the algorithm succeeds in classifying the reads with high accuracy. The probability of assigning the read to the correct class was related to the location in the genome to which the read mapped, which conformed with our expectations since some regions of the genome are more conserved than others. Classifying short reads of high-threat viral pathogens seems to be a promising application of sequence-based deep learning algorithms. Also, recent developments in software and hardware have facilitated the development and training of deep learning algorithms. Further plans of this work are to characterize the hard-to-classify regions of the poliovirus genome, build larger training databases, and expand on the current approach to other viruses.

A65 Characterization of endolysin gene of bacteriophages infecting *Listeria* spp. isolated from dairy industry wastewater

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Bacteriophages and their endolysins, enzymes that degrade the cell walls of bacteria, are emerging as alternative tools to detect and inhibit growth of pathogen bacteria. *Listeria monocytogenes* is a foodborne pathogen that causes listeriosis, a serious invasive disease that affects both humans and a wide range of animals. *Listeria* spp. are ubiquitous in the dairy farm environment and could be present in dairy-processing plants and wastewater. All Listeria-specific bacteriophages found to date are members of the Caudovirales, of the Siphoviridae or Myoviridae families. Myophages infecting Listeria have been recently classified by the ICTV in the Spounavirinae subfamily, as well as in the P100 virus genus. The aim of this work was to isolate *Listeria* spp. bacteriophages and their endolysin codifying genes from wastewater of a dairy industry. Wastewater with and without treatment was sampled during the course of a year, and isolation of bacteriophages was performed after an enrichment step using as hosts *L. innocua, L. ivanovii,* and *L. monocytogenes* serotypes 1/2a, 1/2b, and 4b. Bacteriophages infecting *L. innocua* and *L. ivanovii* were isolated ($n = 24$) from 3 out of 12 samples. Bacteriophages were purified, and the host range was determined using spot test and EOP against five collection strains and several field isolates of *Listeria* spp. Two bacteriophages of narrow and broad host range, vB_Lino_VEfB7, and vB_Liva_VAfA18, were selected for further characterization. High titer stocks of bacteriophages were purified by centrifugation with ammonium acetate, and morphological information on the purified bacteriophages was obtained by negative staining and transmission electronic microscopy. Their morphology, size, and contractile tails indicated that these bacteriophages belonged to the Myoviridae family. Bacteriophage genomes were extracted using phenol-chloroform, followed by ethanol precipitation, and tested by digestion with RNAsa A and DNAse I. RFLP was performed, digesting genomes with restriction enzymes HindIII and NcoI. Consistent with the morphological findings, bacteriophages contained dsDNA genomes but showed different RFLP patterns. A PCR designed to amplify conserved domains of endolysins—PGRP and CwlA—was applied to characterize this gene. Another PCR was designed to amplify the complete endolysin gene, and the complete sequence of this gene was obtained and analyzed. Substitution model selection and a maximum likelihood phylogenetic tree of the endolysin gene was carried out using IQ-Tree software. The sequences of the endolysin gene indicated that the codified enzyme is an N-acetyl-muramoyl-L-alanine amidase, related to A511 and P100 species of the recently described P100virus genus. Further evolutionary analyses are needed to evaluate their belonging to this species or their taxonomy within this genus.

A66 Tracing the evolutionary history of an emerging Salmonella 4,[5],12:i:- clone in the United States

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Salmonellosis is one of the leading causes of foodborne disease worldwide, with an estimated one million cases a year in the United States. Salmonella 4,[5],12: i:-, a monophasic variant of *Salmonella typhimurium*, is an emerging serovar that has been