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Progress toward a deep mutational scanning of the H9 HA to better understand H9N2 avian influenza evolution

Inhalt

Low-pathogenic H9N2 avian influenza is endemic in poultry in many regions of the world, causing significant economic loss and representing a persistent zoonotic threat with pandemic potential as recognized by the WHO. To better understand how each residue of the H9 HA tolerates mutations, we aim to perform a deep mutational scanning (DMS) of its entire coding sequence: this represents a total of 10 659 possible mutations. We have successfully generated a plasmid mutant library with extensive mutational diversity along the H9 gene, that will serve as template for the generation of the mutant virus library. For biosafety consideration, we use a pseudotyped lentiviral system with adapted and optimized conditions to yield high viral H9N2 pseudovirus titers, notably the use of modified 293T as target cells. Finally, a crucial step for DMS experiments is to ensure the genotype-phenotype link when generating the mutant pseudovirus library. Using a previously established two-step protocol to ensure this link, we have successfully rescued mutant H9N2 pseudovirus from cells integrated with lentiviral genomes containing our H9 mutant genes.
These results establish the foundation for subsequent mutations fitness mapping of relevant phenotypic characteristics of the H9 HA such as receptor affinity or antibody selective pressure. Ultimately, DMS data will inform phenotypic assessment of newly collected H9 HA sequences.

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Professional status of the speaker

PhD student

Junior scientist status

Yes, I am a junior scientist.

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