

# Deep mutational scanning of an H3 hemagglutinin can inform evolutionary forecasting of human H3N2 influenza virus

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## Abstract

Abstract text.

## INTRODUCTION

## RESULTS

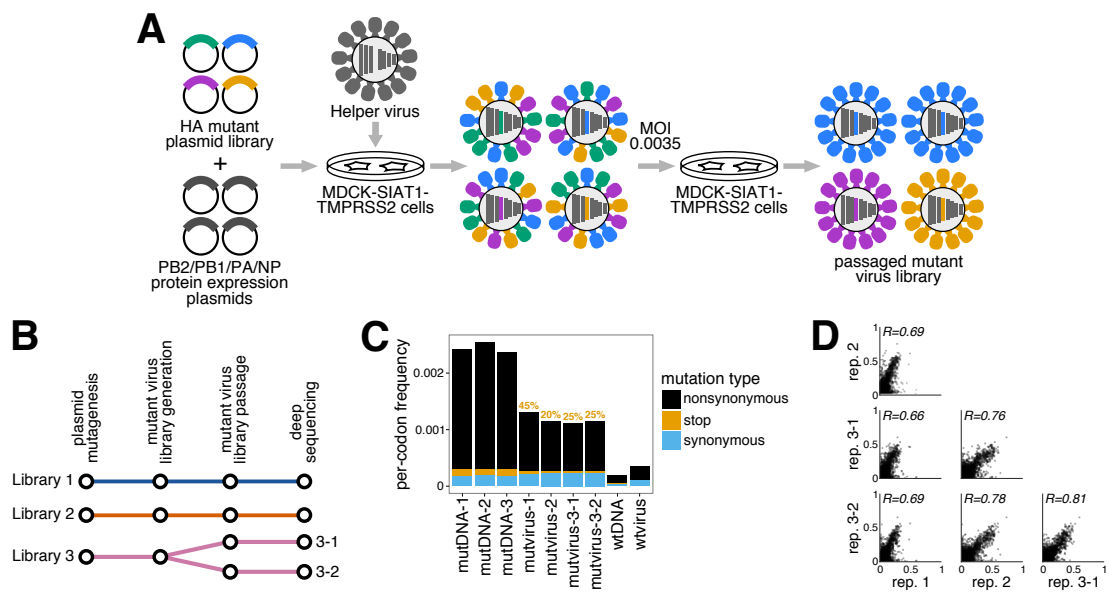
Strategy for deep mutational scanning of an H3 hemagglutinin

H3 site-specific amino-acid preferences

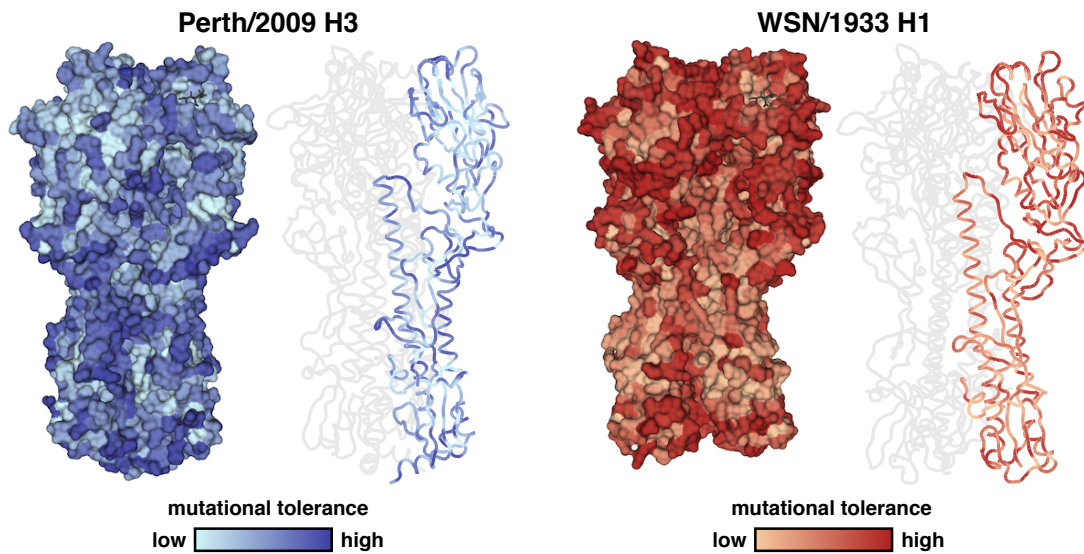
Estimating mutational effects from an H3N2 phylogeny

Comparing H1 and H3 preferences

## DISCUSSION

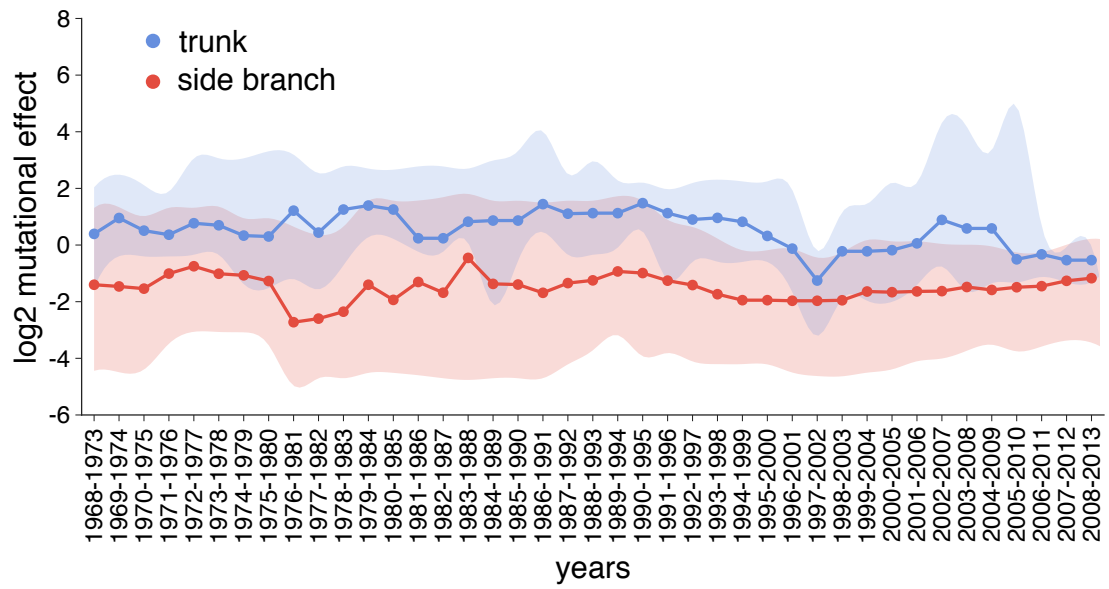


**Figure 1: Overview of deep mutational scanning experiments of H3 hemagglutinin** Figure caption text

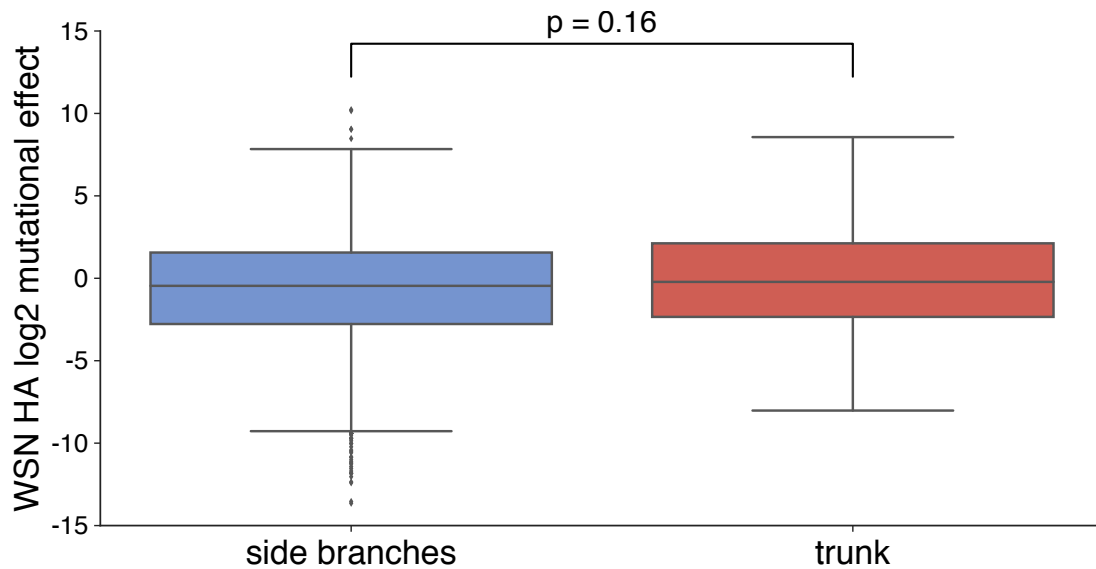


**Figure 2: The mutational tolerance of HA** Figure caption text

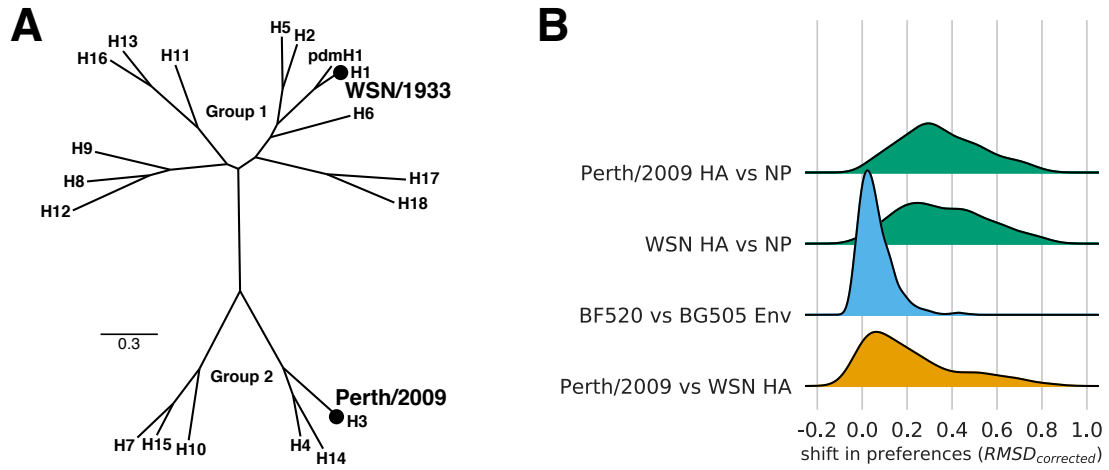




**Figure 5: Sliding window analysis of mutational effects of trunk vs side branches.** Figure caption text



**Figure 6: Trunk vs side branch mutational effects calculated using WSN preferences** Figure caption text



**Figure 7: Distribution of preference shifts for protein homologs** Figure caption text

## **METHODS**

### **HA numbering**

### **Generation of HA codon mutant plasmid libraries**

### **Generation and passaging of mutant viruses**

### **Barcoded subamplicon sequencing**

### **Analysis of deep sequencing data**

### **Quantification of mutational effects and sequence preferences from an H3N2 phylogeny**

### **Data availability and source code**

Deep sequencing data are available from the Sequence Read Archive under BioSample accession [\[\*add accession\*\]](#).

## **ACKNOWLEDGMENTS**

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