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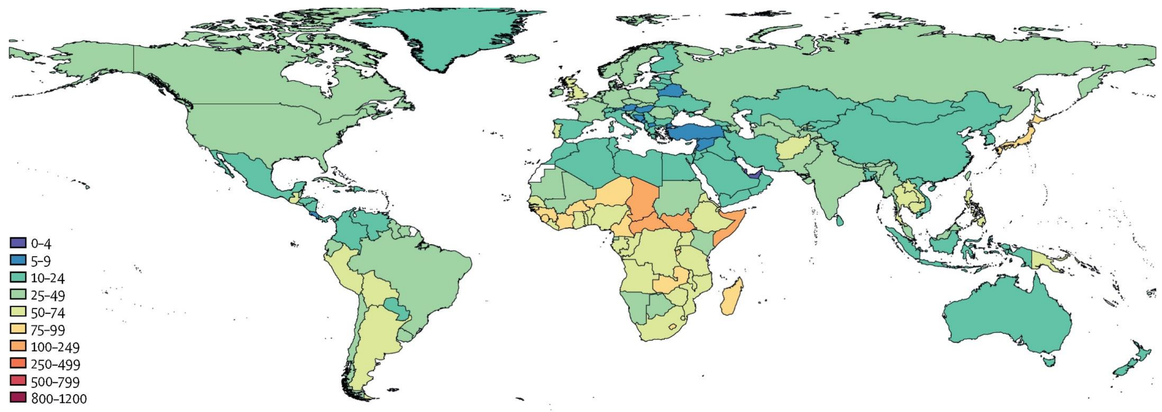
Advantageous Multiplex-NGS Approach for Identifying Whole Genome Respiratory Viruses

Rezak Drali[1], Lionel Chollet[2], Émilie Deroubaix[2], Cecile Poggi[2], Amira Doudou[1], Sofiane Mohamed[1]
[1]ABL Diagnostics, FRANCE; [2]CH - Toulon, FRANCE

Introduction

Acute respiratory infections (ARI) are seasonal infections that cause more than 4.25 million deaths worldwide each year. Their social impact is significant during the onset of epidemics. In this study, we developed an approach based on the simultaneous complete sequencing, using NGS technology, of the genomes of three RNA respiratory viruses namely, SARS-CoV-2, *Influenza* A/B and *Respiratory Syncytial Virus* A/B (RSV).



Global distribution of respiratory infection mortality per 100,000 population.

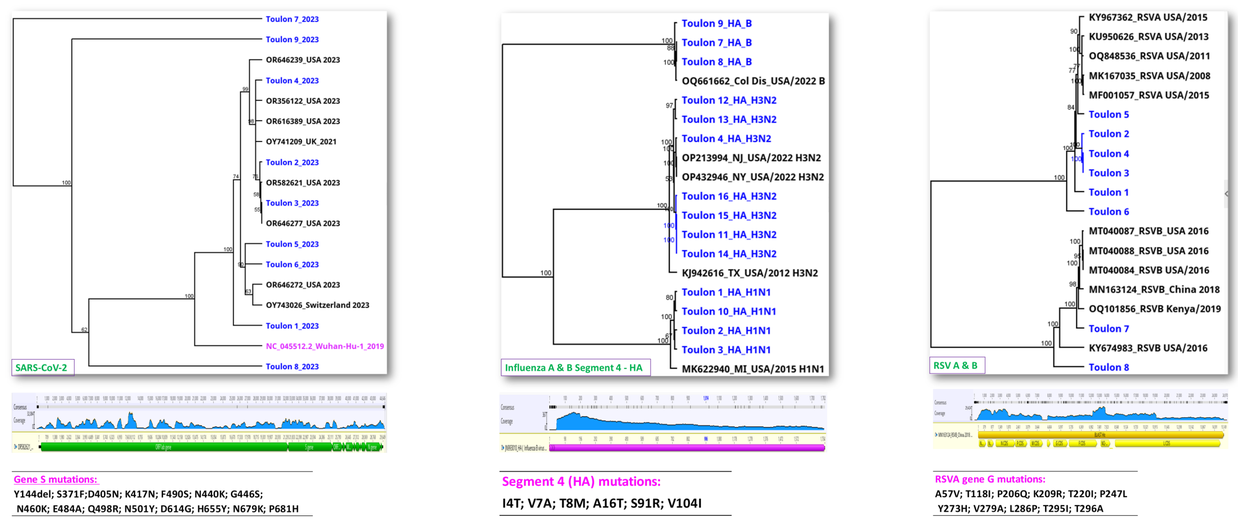
Troeger et al. 2018, The Lancet Infectious Diseases. Volume 18, Issue 11

Methods

Nasopharyngeal specimens of human origin (nine SARSCoV-2, 41 Influenza A/B and eight RSV) were prepared using the QIAamp Mini (Qiagen) kit. The construction of the libraries was carried out with the DeepChek® (ABL) kit. The viruses were sequenced on MiSeq (Illumina). DeepChek® (ABL) software was used to detect mutations and construct phylogenetic trees..

Results

On average, one million reads were generated for each of the sequenced viruses. The quality of the sequencing resulted in a Q30 of 91.1%. Of the nine sequenced SARS-CoV-2 virus samples, six belonged to clade EG.5.1 and three to clade XBB.1.16. Of the influenza virus samples, 26 were *Influenza* A (19 A/H3N2; 7 A/H1N1) and 15 were influenza B/Victoria. Finally, six of the eight RSV viruses sequenced belonged to type A and the remaining two were type B.



Detected mutations and phylogenotyping of respiratory viruses analysed

Conclusions

Multiplex next-generation sequencing of complete genomes of different viruses is an advantageous approach for studies of virus co-circulation and co-infection in pandemic and post-pandemic contexts, as in the case of the respiratory viruses studied here. Indeed, in addition to keeping intact the advantages offered by NGS sequencing technology, namely sensitivity, specificity and precision, this approach also makes NGS sequencing platforms profitable by reducing cost and saving time.

www.linkedin.com/company/abl-sa
Contact information: s.mohamed@ablsa.com