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Do the varying drying rates influence rhinovirus infectivity?

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Book of Abstracts

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The role of proteins in controlling evaporation and hygroscopic behavior of exhaled respiratory droplet

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Background and Objective:
The transmission of respiratory pathogens via exhaled droplets and aerosols is a key mechanism in the spread of infectious diseases such as influenza and COVID-19. The physicochemical properties of exhaled respiratory droplets, which vary under different environmental conditions, directly influence the survival and transport of airborne pathogens. Factors such as temperature, relative humidity (RH), sunlight, and pH play crucial roles in determining pathogen activity. Among these, RH has been shown to significantly impact pathogen activity in respiratory droplets; however, the underlying mechanisms remain unclear, particularly given the complex composition of respiratory droplets. A deeper understanding of the physicochemical properties of exhaled respiratory droplets is essential for comprehending the spread of airborne diseases and predicting aerosol transmission dynamics. In this study, we investigate the role of mucin and albumin, key respiratory proteins, in controlling droplet evaporation and hygroscopic behavior, with the aim of better understanding RH-dependent pathogen activity within respiratory droplets.
Methods:
An electrodynamic balance (EDB) was used to levitate individual droplets under RH conditions ranging from < 5% to 97%. This setup prevented the droplets from contacting any surface, enabling measurements of their evaporation and the hygroscopic growth. Changes in droplet mass and size were measured using the two-dimensional light scattering patterns recorded during the experiment. The morphology and chemical composition of droplets in their equilibrium state were analyzed using an environmental scanning electron microscope (ESEM) equipped with an energy-dispersive X-ray (EDX) spectrometer, with 50 Pa N2 as the background gas.
Results:
Our results show that the presence of mucin affected the evaporation and the rehydration of the respiratory droplets. Specifically, mucin was found to slightly retard the evaporation process, which is likely due to the formation of a semi-solid layer on the surface of the droplets during evaporation. This layer appears to impede the uptake of water, thus influencing the hygroscopic growth behavior of the droplets. While the results for mucin are conclusive, data analysis for albumin is still ongoing, and further insights into its role in droplet behavior will be presented once the analysis is completed. However, SEM images suggest that , unlike mucin, albumin does not form a dense outer shell under dry conditions, highlighting a distinct difference in the behavior of these two proteins.
Conclusion:
Our results highlight the significant role of different proteins in affecting the properties of exhaled respiratory droplets during evaporation and rehydration. The organic content of respiratory fluids varies depending on the region of the respiratory tract where it is produced. Virus-laden respiratory droplets generated in areas with higher organic content may form a more robust shell under dry conditions, thereby enhancing the virus's environmental survivability by protecting it from factors such as temperature, humidity, and ultraviolet radiation. The variation in the physicochemical properties and morphology of respiratory droplets, especially in the presence of different organic compounds and varying organic content, remains an area worthy of further exploration and should be the focus of future research.

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Do the varying drying rates influence rhinovirus infectivity?

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Objective: Human rhinovirus is the most common cause of the common cold worldwide (1). It has been shown that a substantial part of the airborne viruses is found in aerosol particles in the range of 1-4 μm (2). Studying the infectivity of aerosol particles in this range is, however, challenging; therefore, not many studies have been conducted on their infectivity. The aim of this work is to investigate the infectivity of aerosolized rhinovirus in particles <5 μm under varying levels of relative humidity (7%, 50%, and 80%-90%).
Methods: We performed aerosolization and collection of rhinovirus in a laboratory setup previously described by Alsved et al. (3). A flow tube was placed inside a laminar flow (LAF) cabinet to avoid any contamination during the experiment. The BioAerosol Nebulizing Generator (BANG) was used to generate the aerosol of rhinovirus, which was introduced into either a long or short exposure tube under different levels of relative humidity (RH). At the other end of the exposure tube, the bioaerosol was collected by impaction in three different size fractions using the BioCascade (Aerosol Dynamics Inc.): >10 μm, 4-10 μm and 1.5-4 μm. The remaining particles <1.5 μm continued to the BioSpot-VIVAS (Aerosol Devices) where they were grown to larger droplets by water condensation before impaction into liquid. In addition, an aerodynamic Particle Sizer (APS, Model 3321, TSI Inc.) and a Scanning Mobility Particle Sizer (SMPS, TSI Inc.) were used for analyzing the size distribution of the bioaerosol. To ensure that we were measuring the dry size of the particles, a silica drier was connected before the APS and SMPS. Additionally, the viral load of the collected bioaerosol samples was determined by quantitative polymerase chain reaction (qPCR). Since qPCR only detects the total presence of cDNA in a solution and does not assess the infectivity of the virus, the infectivity of rhinovirus was assessed by measuring the cytopathic effect in HeLa cells, using the 50% Tissue Culture Infectious Dose (TCID50) and the Most Probable Number (MPN) method. To minimize the influence of small variations in aerosol concentration on virus infectivity results, MPN values were normalized by the total aerosol mass measured by the APS during the sampling time.
Result: In the experiment when all particle sizes were collected with the BioSpot, our results suggest that airborne rhinovirus infectivity was about 50% higher at RH above 80% compared to a 7% RH, however, it was not statistically significant. When collecting the aerosol in different size fractions using the BioCascade and the BioSpot, the smallest particle size fraction (<1.5 μm) was significantly more infectious than the two larger size fractions (1.5-4 and 4-10 μm) when aerosolized at 7% RH (t-test, $p<0.05$). No difference in infectivity was found when comparing larger particles to each other (4-10 μm vs 1.5-4 μm). The infectivity of the largest particle size fraction (>10 μm) was below the detection limit of the MPN assay.
Conclusion: Based on the experimental results, aerosol at high humidity and particles smaller than 1.5 μm contained more infectious rhinovirus per aerosol mass than aerosol in low humidity and in particles >1.5 μm. There is a possibility that the collection methods, direct impaction for particles >1.5 μm versus condensational growth prior to impaction for <1.5 μm, influenced the result. So far, experiments have only been conducted once, so repeating the experiment is essential to be able to draw any firm conclusions. In addition, we will develop a copy standard for the qPCR to be able to normalize the infectivity by the virus copy number.
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Modelling indoor NO2 exposures to enable health impact assessment of gas cooking emissions

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