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CONTROLLING THE SIZE DISTRIBUTION OF EXHALED BIOAEROSOL DROPLETS BY MODULATING THE VISCOELASTIC PROPERTIES OF HUMAN AIRWAY MUCUS

Md Anwarul Hasan[1], Carlos F. Lange[1] and Malcolm King[2]

[1]Department of Mechanical Engineering, University of Alberta, Canada
[2]Pulmonary Research Group, Department of Medicine, University of Alberta, Canada

ABSTRACT

Effect of the viscoelastic properties of artificial mucus simulant samples on the volume size distribution of bioaerosol droplets generated during simulated coughing has been investigated through in-vitro experiments. The mucus simulant samples had similar viscoelastic properties as real human airway mucus. The mucus simulant gels were prepared by mixing various proportions of 1% Locust bean gum solution and 0.1 M sodium tetraborate (XLB) solution. The viscoelastic properties of the samples were measured using a Bohlin Gemini (Malvern) nano rheometer with peltier plate assembly. An artificial cough machine was used to simulate human cough, generating aerosol droplets in a model trachea attached to the front of the cough machine. The size distribution of the droplets generated through simulated cough was measured using a laser diffraction particle sizer (Malvern SprayTec). Results confirm that the viscoelastic properties of mucus have substantial effect on the size distribution of bioaerosol droplets generated during coughing. The experimental results showed an increase in particle size as the sample changed from an elastic solid type to a viscoelastic type to viscous fluid type sample.

Keywords: Bioaerosol droplets, Mucus, Viscoelasticity, Surface Tension, Cough machine.

1. INTRODUCTION

Bioaerosol droplets generated and exhaled by humans during breathing, sneezing and coughing may carry airborne pathogens, which upon inhalation by others spread certain contagious infectious diseases, such as influenza, measles, swine flue, chickenpox, smallpox, SARS and so on. Such airborne transmission of respiratory infections through the bioaerosol droplets poses a major public health threat and it is a subject about which surprisingly very little is known. Recent theoretical studies [1] support the hypothesis that the number and size distribution of the bioaerosol droplets generated during coughing can be controlled by altering viscoelastic and surface properties of the airway surface liquid. However, the detail about the correlations between the viscoelastic properties of mucus and the characteristics of the generated bioaerosol droplets are still unknown. In order to mitigate the airborne transmission and to control the generation of bioaerosol droplets during coughing, sneezing etc., it is important to understand the detail mechanisms underlying these events (coughing, sneezing etc) as well as the structure and properties of the airways surface mucus, and the correlations between the relevant viscoelastic properties of mucus with the size distribution of the generated droplets.

1.1 State of the Art Knowledge about Coughing and Bioaerosols

A cough can generate some 3000 droplet nuclei, as can talking for 5 minutes [2]. A sneeze can generate as many as 40,000 droplets [3]. IRSST - Guide on respiratory protection against bioaerosols [4] reports these numbers to be even higher, according to which, during a sneeze, close to two million droplets can be expelled at a velocity of 100 m/sec (200 miles/hour), compared to fewer than 100,000 droplets from a cough. This significant difference is based on the origin of the secretions, which is deeper in the case of a cough [5]. During a forceful cough airspeeds as high as 200 m/s can be attained [6]. Many of the generated droplets may be large enough to contain thousands of microorganisms [7].

During the expulsion, the diameters of the droplets vary between 1 and 2,000 μm, 95% of which are in the order of 2 to 100 μm. However, they dry very rapidly. The drying times for 100 and 50 μm droplets in air at 50% relative humidity are 1.3 and 0.3 seconds, respectively [8]. After being exhaled these droplets can evaporate to particles in the size range of 0.5 to 12 micron [3]. Others reported that the infectious particles generated from human respiratory sources occur primarily as droplet nuclei of 0.5 to 5.0 micron in

diameter [9]. On complete evaporation, the particles may be small enough to remain airborne in the indoor air flow.

The size of microorganisms, on the other hand, span wide size ranges i.e. from 0.3 to 10 micron for bacterial cells and spores, 2.0 to 5.0 micron for fungal spores, and 0.02 to 0.30 micron for viruses [10]. Others report these numbers to be slightly different. For example: According to Lee et al. [11], most bacteria and molds are between 0.7 and 10 μm in size. According to AIHA (American Industrial Hygiene Association) the sizes of bioaerosols are in the order of 0.02 to 0.25 μm for viruses, 0.3 to 15 μm for bacteria, and 1 to 50 μm for the majority of molds and yeasts [12]. Yassi and Bryce [5] and ACGIH [7] report that the size of infectious bioaerosols is probably between 0.1 and 10 μm [5], [7]. It even appears that the majority of viruses and bacteria that cause respiratory diseases in humans are usually inside bioaerosols with diameters greater than 5 μm.

Depending on aerosolized droplet size, airborne pathogens can quickly be deposited on nearby external surfaces or the expired bioaerosols can travel great distances and remain airborne for an extended period of time, particularly when droplet diameters are too large for diffusive deposition (>200 nm) or too small for gravitational deposition (<2 μm) [13, [14]. Bioaerosols consisting of solid or liquid particles smaller than 10 μm in diameter remain suspended in the air for a sufficient time (a few hours) and are likely to be inhaled [7], [15], [5]. Table 1 presents the time required for a bioaerosol to be deposited by sedimentation from a height of three meters. As shown in table 1, solid or liquid particles between 6 and 10 μm can take a few hours before being deposited from a height of 3 meters [8], [5].

Some researchers make a distinction between micro droplets with diameter less than or equal to 5 micron, and those with diameter > 5 micron, with a belief that droplets larger than 5 micron sediment before traveling a distance of one meter. Such a distinction has no solid foundation (Lenhart et al., 2004b).

Table 1: Behavior of bioaerosols in the air (Yassi and Bryce, 2004)

Diameter in micron	Time required for deposition from a height of 3 meters
100	10 sec
40	1 min
20	4 min
10	17 min
6 to 10	A few hours
0.06 to 6	Several hours

1.2 Literature Review on Bioaerosol Research

To date, few studies have carefully examined the nature of the bioaerosols that humans exhale on a daily basis (Table 1). Early researchers assumed the upper respiratory tract (nose, mouth and throat) was the primary location of droplet formation [16], [17], [18]. In these early studies, the mouths and throats of volunteers were coated with a dye and breathing, talking, sneezing and coughing maneuvers were monitored and any resulting droplets were collected directly onto a slide. Only droplets >1 μm were measured by microscopic observation. Duguid [16] found that droplets (>1 μm) produced by speaking, coughing and sneezing were sufficiently small enough to remain airborne. Normal breathing, however, produced no measurable droplets (>1 μm). In a second series of experiments Duguid [17] determined that coughing produced an average droplet size of 14 μm and geometric standard deviation (GSD) = 2.6 and for sneezing GM = 8.1 lm and GSD = 2.3. On the other hand, Loudon and Roberts [18] showed that the estimated lognormal parameters were GM = 12.1 m and GSD = 8.4 for cough. More recently, Papineni and Rosenthal [19] measured expired bioaerosol droplets (in nose and mouth breathing, coughing and talking) to be <2 lm in size, with no droplets >8 lm.

An interesting common finding from these studies was the high variability in the levels of bioaerosol production from different individuals. More-recent experiments have utilized optical particle counting (OPC) to determine the size and concentration of droplets exhaled from all parts of the respiratory tract [19, [20], [21]. In a similar experiment to that of Papineni and Rosenthal, Edwards et al. [20] observed 11 healthy human subjects. Results from this study confirmed Papineni and Rosenthal's findings because they suggested exhaled particles during normal mouth breathing are predominantly <1 μm in diameter. Edwards' results also showed that expired particle numbers vary substantially from subject to subject, with two distinct populations: low producers (those exhaling an average of <500 droplets per liter over a six hour measurement period) and super producers (those exhaling an average of >500 droplets per liter over a six hour measurement period) of expired bioaerosols. Remarkably, the super producers (six people from this test group) expired 99% of the total amount of bioaerosols that were expired by the entire group,

In their study, Edwards et al. [20] further found that delivering ~1 g of isotonic saline (orally via nebulized aerosols, 5.6 μm in diameter) reduces the total amount of expired aerosols (among the super-producing individuals) by ~72% over a six hour period and markedly diminishes total expired bioaerosol production for the entire group. In vitro results, obtained using a simulated cough machine, also indicated that a mucus mimetic nebulized with saline produces a larger droplet size after the forced convection of air over its surface than when air is forced over the mucus mimetic alone (i.e. without saline nebulization). These results led Edwards and co-workers to conclude that saline delivered onto lung surfactant increases its surface tension, and potentially other dynamic physical properties of the lung surfactant, thereby changing the droplet breakup dynamics. In another study, Clarke et al. [22] report that delivering isotonic saline aerosols (in 5.6 μm droplets) into the endotracheal tube of anesthetized bull calves showed a dose-responsive effect on exhaled bioaerosols; six minutes of treatment resulted in a decrease ≤ 50% of exhaled bioaerosols for at least 120 minutes, compared with pre-treatment.

The idea of inhaling saline for medical benefit has existed since the time of Hippocrates. However, the mechanism of this phenomenon remains unclear, as does the reason for the dramatic exhaled aerosol differences among human individuals [23].

In explaining why certain individuals in Edward's study breath out many more bioaerosol particles than do others, Wiwik et al suggested that possibly transient and/or durable intersubject variations in the ionic composition of airways surface liquid produce the intersubject variations in the number of expired bioaerosol particles, and potentially in airborne disease infectiousness. They reasoned that the gelation of the free surface of airways surface liquid mimetic owing to deposition of salt-based solutions should diminish the propensity of the airways surface liquid to break up into droplets in the cough machine.

2. EXPERIMENT

2.1 Material and Specimen

Mucus simulant gels having similar viscoelastic properties as real human airway mucus [24] over a wide range were used. The gels were prepared by mixing various proportions 1% Locust bean gum (LBG) solution and 0.1 M sodium tetraborate (XLB) solution. For preparation of locust bean gum solution, 100 ml of saline (Na_2NO_3) solution was heated to 80 ^{0}C. The required amount of LBG powder was added to the saline solution little by little while the solution continuously being stirred using a magnetic stirrer. The weight of the beaker with the saline solution was measured both before and after heating and mixing LBG powder. Mass balance was ensured by adding additional saline solution to compensate the losses due to evaporation whenever necessary. Compositions of locust bean gum solution used in the experiments was 1%. 0, 10, 15, 20, 30, 45, 60 and 90 microlitre of 0.1M tetraborate solution was added to 1.5 ml of locust bean gum solution. The mixture was then vortexed for 2 minutes on a vortex meter.

2.2 Viscoelastic Properties

Viscoelastic properties of the samples were measured using a Bohlin Gemini Malvern Nano-Rheometer with a peltier plate assembly and parallel plate geometry. The frequency of the oscillation was varied between 0.1 Hz and 10 Hz. All measurements were performed at constant strain within the linear viscoelastic region i.e. within the limit of shear stress where viscosity is independent of applied shear stress. This region was obtained for each sample by performing an amplitude sweep test before the actual measurement of viscoelastic properties. The temperature was kept constant at 25 ^{o}C. A solvent trap was used to avoid drying out of the samples during measurement.

2.3 Cough machine experiment

Cough machine experiments were performed to mimic bioaerosol generation during a human cough. The artificial cough machine comprised of an 8-liter Plexiglas tank equipped with a Wilmot Castle pressure gauge. The Gas release was controlled by an Asco solenoid valve located at the start of the outflow line. A model trachea 35 cm long with an interior width of 2.0 cm and an interior height of 1.0 cm was attached to the outlet after the solenoid valve. A small rectangular slot 3 cm long by 1.76 cm wide and 1 mm deep was engraved on the upper surface of the bottom plate of the model trachea to hold the mucus simulant sample in place. The slot size was large enough for holding 1.5 ml sample. Mucus simulant samples were placed inside the trachea at the sample slot and a normal adult cough was simulated by suddenly releasing the air from the tank stored at a pressure of 8.5 psi.

2.4 Measurement of Droplet Size Distribution

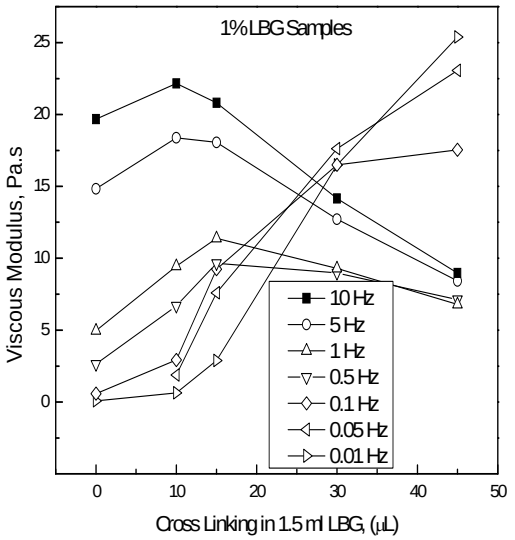
The size distribution of the droplets was measured using a laser diffraction based size measuring device, Malvern Spraytec. In our case, the droplets generated from the cough machine were passed through the laser beam of of about 1cm diameter. The droplets scatter the light, smaller droplets scattering the light at larger angles than bigger droplets. The scattered light is measured by a series of photodetectors placed at different angles. This is known as the diffraction pattern for the sample. The diffraction pattern is used to measure the size of the droplets using light scattering theory developed by Mie. Laser diffraction is sensitive to the volume of the droplets. For this reason, droplet diameters are calculated from the measured volume of the droplets, assuming a sphere of equivalent volume.

3. RESULTS AND DISCUSSION

The viscous modulus and elastic modulus versus amount of cross linking graphs for the mucus simulant samples are shown in Fig 1 (a) and (b) respectively. The Figure shows that the change in viscous modulus with increase in cross linking follows different pattern at different frequencies (i.e. at different shear rate). Even though at low frequencies ($\leq$0.1 Hz) the change in viscous modulus with increase in cross linking is considerably high, at moderate to high frequencies ($\geq$ 0.5 Hz), which are more likely relevant to coughing, change in viscous modulus with increase in cross linking is not significant. The elastic modulus of the samples, on the other hand increases continuously by up to two orders of magnitude as shown in Fig. 1(b). Figs. 2 (a) and (b) present the complex viscosity of the samples against frequency and amount of cross linking respectively. The shear thinning behavior is evident in Fig. 2 (a) while Fig. 2 (b) shows that the complex viscosity increases with increase in cross linking at all frequencies. Figs. 3 (a) and (b) show the phase angle of the samples against frequency and amount of cross linking respectively. As expected due to the non-Newtonian behavior of the samples, the phase angle values are different at different shear rate for the same sample. It is worth mentioning here that a zero degree phase angle means a pure elastic (solid) material, a ninety degree value of phase angle indicates a purely viscous (Newtonian fluid) material while the values between zero and ninety indicates a viscoelastic material. Based on the phase angle at a particular shear rate, the samples can be classified into (i) elastic solid like samples (0 to <30 degree), (ii) strongly

viscoelastic samples (30 to 60 degree), and (iii) viscous liquid like samples (60 to 90 degree). A simulated Cough is considered to be a high shear rate phenomenon as is a real human cough considered. However since the actual value of the shear rate that governs the droplet generation is unknown, for convenience we will subsequently refer all viscoelastic properties at a frequency of 0.1 Hz. Thus based on the phase angle values at 0.1 Hz, from Fig. 3 (a) we find that 0 micro liter sample was a viscous fluid type sample, 10 micro liter and 15 micro liter samples were strongly viscoelastic samples while the 30 and 45 micro liter samples were elastic solid type samples.

The volumetric size distribution of the droplets generated during simulated cough experiments for addition of 0, 30, 45 and 60 micro liter cross linking agent (Sodium Tetraborate solution) are presented in



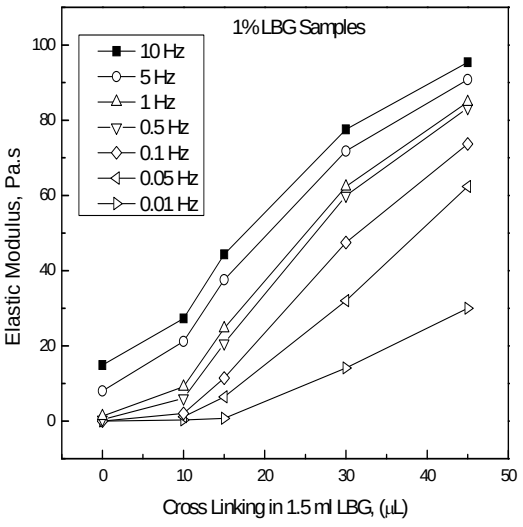


Fig 1. Viscous modulus and elastic modulus versus amount of cross linking graphs for 1%LBG samples.

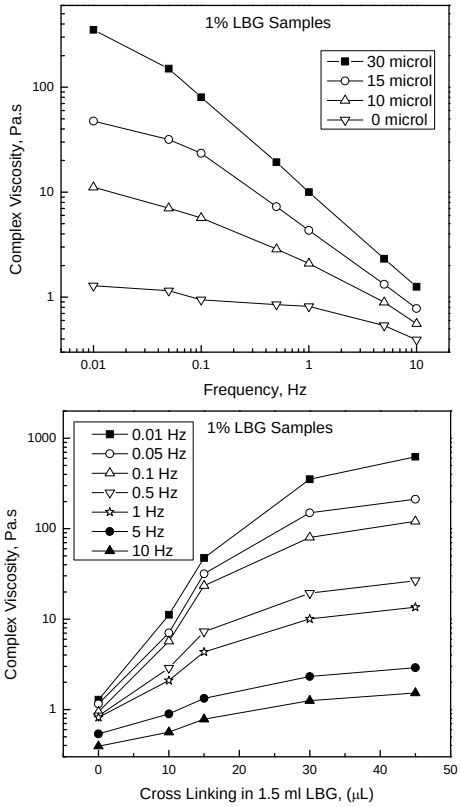


Fig 2. Complex viscosity versus (a) frequency, and (b) amount of cross linking graphs for 1%LBG samples.

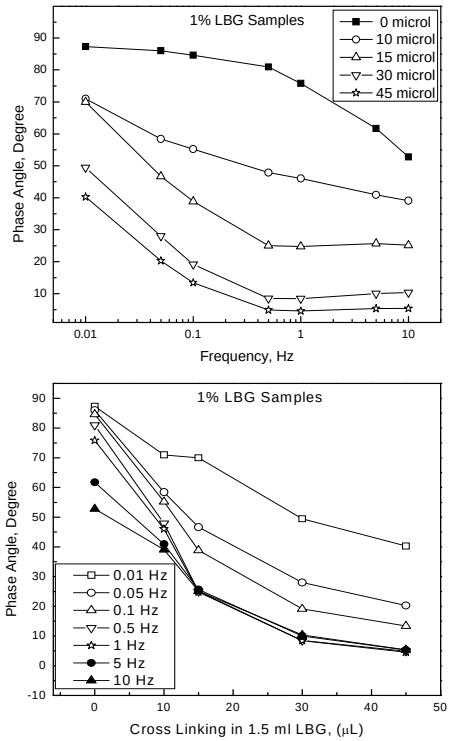


Fig 3. Phase angle for 1%LBG samples.

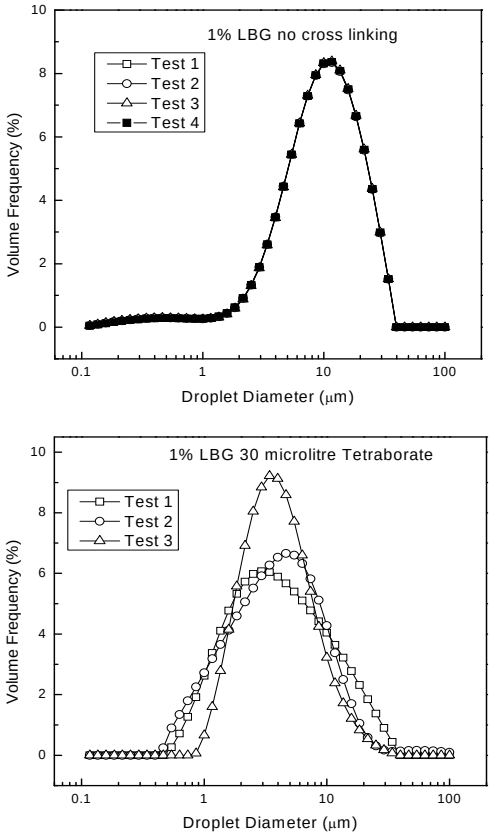
Figs. 4 (a), (b), (c), and (d) respectively. Each figure presents results for at least three repeated test runs for one sample. Figure 4 confirms that the viscoelastic properties of mucus have substantial effect on the size distribution of droplets generated during coughing. Three

distinct type of droplet size distribution are apparent in Figure 4. While the viscous liquid like sample, Fig. 4(a), shows a bimodal droplet size distribution with a smaller peak at around 0.4 micron and a larger peak at around 10 micron, the viscoelastic type sample, Fig. 4 (b), shows a single mode of size distribution. The smaller peak in the submicron size range apparent for the viscous liquid like sample is no longer apparent for strongly viscoelastic sample. For elastic solid type samples, Figs. 4 (c) and (d), the size distribution is again a bimodal distribution with a much stronger peak at a relatively smaller size.

In another word, the size distribution of the droplets apparently shifts rightward as the sample changes from an elastic solid like sample (large amount of cross linking) to a viscoelastic sample to a viscous fluid like sample as shown in Fig. 5.

4. CONCLUSIONS

This study confirms that the viscoelastic properties of mucus have substantial effect on the size distribution of droplets generated during coughing. The experimental results showed an increase in particle size as the samples changed from an elastic solid type to a viscoelastic type to viscous fluid type samples.



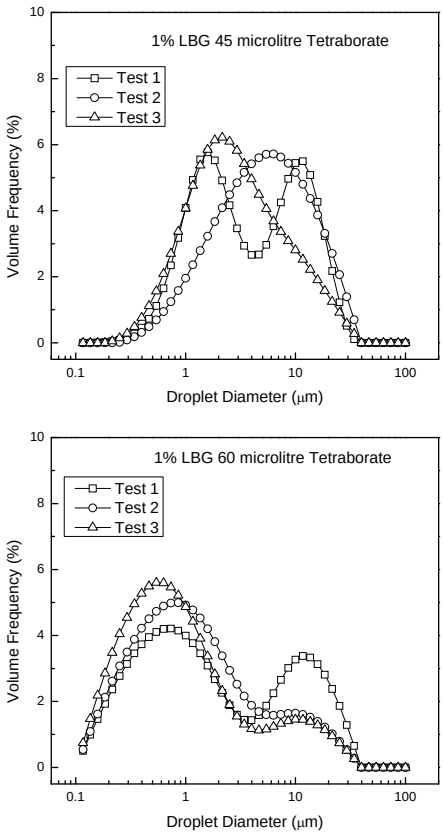


Fig 4. Complex viscosity versus (a) frequency, and (b) amount of cross linking graphs for 1%LBG samples.

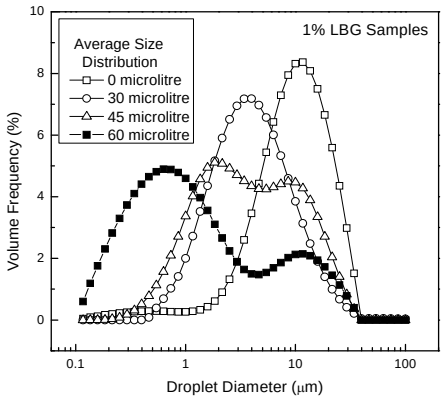


Fig 5. Complex viscosity versus (a) frequency, and (b) amount of cross linking graphs for 1%LBG samples.

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6. REFERENCES

1. Vasudevan M., and Lange C. F., 2007, "Surface Tension Effects on Instability in Viscoelastic Respiratory Fluids", Mathematical Biosciences, 205**:** 180-194.
2. Des Prez R., and Heim C., 1990. Mycobacterium Tuberculosis. In: Mandell, G., Douglas, J. R., and Bennett, J., (Eds.), Principles and Practices of Infectious Diseases. Churchill Livingstone, New York, pp. 1877-1906.
3. Cox C. S., 1987. The Aerobiological Pathway of Microorganisms. John Wiley and Sons Ltd, Chichester (UK).
4. Lavoie J., Cloutier Y., Lara J., and Marchand G., 2007. Guide on Respiratory Protection against Bioaerosols - Recommendations on Its Selection and Use, Studies and Research Projects/Technical Guide RG-501 IRSST, Montréal, pp. 40 pages.
5. Yassi A., and Bryce E., 2004, "Protecting the Faces of Health Care Workers: Knowledge Gaps and Research Priorities for Effective Protection against Occupationally-Acquired Respiratory Infectious Diseases. The Change Foundation, Ontario Hospital Association and Occupational Health and Safety Agency for Healthcare in Bc, 103 Pages".
6. Ross B. B., Gramiak R., and Rahn H., 1955, "Physical Dynamics of the Cough Mechanism", J. Appl. Physiol., 8**:** 264–269.
7. ACGIH, 1999. American Conference of Governmental Industrial Hygienists (Acgih), (1999), Bioaerosols: Assessment and Control. , Cincinnati, OH, pp. 322 pages.
8. Lenhart S. W., Seitz T., Trout D., and Bollinger N., 2004b, "Issues Affecting Respirator Selection for Workers Exposed to Infectious Aerosols: Emphasis on Healthcare Settings ", Applied Biosafety, 9**:** 2-36.
9. Owen M. K., Ensor D. S., and Sparks L. E., 1992, "Airborne Particle Sizes and Sources Found in Indoor Air", Atmos. Environ., 26**:** 2149-2162.
10. Cole E. C., and Cook C. E., 1998, "Characterization of Infectious Aerosols in Health Care Facilities: An Aid to Effective Engineering Controls and Preventive Strategies", American Journal of Infection Control, 26**:** 453-464.
11. Lee S.-A., Adhikari A., Grinshpun S. A., McKay R., Shukla R., Zeigler H. L., and Reponen T., 2005, "Respiratory Protection Provided by N95 Filtering Facepiece Respirators against Airborne Dust and Microorganisms in Agricultural Farms. " Journal of Occupational and Environmental Hygiene, 2**:** 577-583.
12. Goyer N., Lavoie J., Lazure L., and Marchand G., 2001. Les Bioaérosols En Milieu De Travail : Guide D'évaluation, De Contrôle Et De Prévention. Études Et Recherches, Guide Technique T-23,. Institut de recherche Robert-Sauvé en santé et en sécurité du travail, pp. 63 pages.
13. Gerrity T. R., 1979, "Calculated Deposition of Inhaled Particles in the Airway Generations of Normal Subjects", J. Appl. Physiol., 47**:** 867–873.
14. Stahlhofen W, Gebhart J, Heyder J, Philipson K, and P. C., 1989, "Intercomparison of Experimental Regional Aerosol Deposition Data", J. Aerosol Med. , 2**:** 285–308.
15. Hinds W. C., 1982. Aerosol Technology. Properties, Behavior, and Measurement of Airborne Particles. John Wiley and Sons, Toronto.
16. Duguid J. P., 1945, "The Numbers and Sites of Origin of the Droplets Expelled During Expiratory Activities", Edinburgh Med. J., 52**:** 385-401.
17. Duguid J. P., 1946, "The Size and Distribution of Air-Carriage of Respiratory Droplets and Droplet-Nuclei", J. Hyg., 4**:** 471–480.
18. Loudon R. G., and Roberts R. M., 1967, "Droplet Expulsion from the Respiratory Tract", American Review of Respiratory Disease 95**:** 433–442.
19. Papineni R. S., and Rosenthal F. S., 1997, "The Size Distribution of Droplets in the Exhaled Breath of Healthy Human Subjects ", J. Aerosol Med., 10 105–116.
20. Edwards D. A., Jonathan C. Man, Brand P., Katstra J. P., Sommerer K., Stone H. A., Nardell E., and Scheuch G., 2004. Inhaling to Mitigate Exhaled Bioaerosols, Proc. Natl. Acad. Sci. U. S. A. , pp. 17383–17388.
21. Fairfield C. I., and Stampfer J. F., 1987, "Particle Concentration in Exhaled Breath", Am Ind Hyg Assoc J. , 48**:** 948–949.
22. Clarke S. W., Jones J. G., and Oliver D. R., 1970, "Resistance to Two-Phase Gas-Liquid Flow in Airways", J. Appl. Physiol., 29**:** 464-471.
23. Watanabe W., Thomas M., Clarke R., Klibanov A. M., Langer R., Katstra J., Fuller G. G., Griel L. C., Fiegel J., and Edwards D., 2007, "Why Inhaling Salt Water Changes What We Exhale", Journal of Colloid and Interface Science, 307**:** 71-78.
24. King M., Brock G., and C. Lundell C., 1985, "Clearance of Mucus by Simulated Cough", J Appl. Physiol, 58**:** 1776-1982.

7. MAILING ADDRESS

Md Anwarul Hasan
Department of Mechanical Engineering, University of Alberta, Canada.