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Bioaerosols and Corona Virus Diffusion, Transmission, Carriers, Viral Size, Surfaces Properties and other Factor Involved



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ABSTRACT

Aim of this work is to evaluate the chemical-physical binding between some respiratory virus and their carrier like particulate matter, water particles dust and other. This make possible to verify if a virus is airborne or not and the forces that regulate the bioaerosol containing virus. The corona-virus envelope whit their chemical -physical properties and electrical charge are fundamental to fully understand properties, diffusion pattern, surviving and other relevant in spread of this respiratory disease. Medicine, biology but also chemistry and physics can help in fight with such Pandemic disaster. The result of this study are then useful in preventing strategy by public international health institution.

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1. Introduction

Since the first stages of life evolution In the earth bio-aerosols was an crucial environment. This kind of environment follow chemical-physics rules and are subjected to perturbation of this property. This bio-aerosols contain various form of life sine also bacteria , fungi but also virus. Actual Covid-19 pandemia must to be analyzed under this point of view: An bioaerosols that follow chemical-physics law.

The rapid and logarithmic explosion of cases in the second wave in France, UK, Spain in a few time (weeks) seem to show that not only transmission. By direct contact and by droplets: also airborne must to be deeply evaluated. The lower PM particle in example penetrate inside deeply in the lungs then higher and this factor can be related to the severity of the disease.

But this chemical physics properties of the link between virus and carrier in Aerosol are correctly taken in consideration in the preventing strategies? Chemical physic properties of virus envelope can be responsible of the link with carrier but also to repulsion between virus increasing Brownian moto.

According Daniel Verreault et al : related the methods for sampling of airborne viruses:

"Any microorganism, including viruses, can become airborne. Contaminated material can be aerosolized in many different ways, ranging from wind to human and animal activities such as sneezing, mechanical- processes, etc. If the aero-dynamic size of an infectious particle is appropriate, it can remain airborne, come into contact with humans or animals, and potentially cause an infection. The probability of an airborne micro-organism-laden particle causing an infection depends on its infectious- potential and its ability to resist the stress of aerosolization. Many epidemiological- studies have proposed that viruses can spread from one host to another by using air for transport. The capacity of the foot-and-mouth disease (FMD) virus to spread by air has been studied and reviewed over the years and is now being investigated using computer- models. One of these models predicted that in a "worst-case scenario" of an FMD outbreak, cattle could be infected as far as 20 to 300 kilometers away from an infectious -source . Dispersion- models based on meteorological -data and information on the spread of FMD at the beginning of the 1967-68 epidemic in the UK strongly suggested that the infection may have spread by the airborne- route over a distance of 60 km . Airborne- transmission of FMD was also reported to have occurred during the 1982-1983 epidemic in Denmark. In the latter

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case, an analysis of epidemiological dynamics using molecular- methods coupled with meteorological data concluded that the infection had spread by air over a distance of 70 km . Similarly, the results of a Canadian study on an FMD epidemic reported that airborne- viruses may have traveled 20 km down-wind from the contaminated source . a recent study on the O/UKG/2001 strain of FMD virus indicated that it does not spread efficiently between sheep by the airborne- route.

Other strains may behave differently. Airborne micro-organisms can represent major health and economic- risks to human and animal -populations. Appropriate preventive actions can be taken if the threat posed by such micro-organisms is better understood. Authorities need to be aware of the nature, concentration, and pathogenicity of airborne-micro-organisms to better control them. This information can be obtained by using various air sampling -methods, each of which has its particular advantages and disadvantages. Many types of samplers and analytical methods have been used over the years.

A virus can multiply only within a host- cell. Infected cells can spread viruses directly into the surrounding- air (primary-aerosolization) or to fluids and surfaces, which can become sources for airborne- transmission (secondary-aerosolization). Secondary-aerosolization can occur for any virus, predominantly when air displacements or movements around contaminated surfaces or fluids disperse the viruses into the air. It can also occur by liquid- splashes, which can aerosolize viruses in liquids or on surfaces. Almost any kind of disturbance of infected organisms or materials, even the bursting of bubbles in sea-water, can produce airborne, virus-laden particles. Farm-animals have also been studied for their emission of airborne -viruses. The FMD virus, which is one of the most widely studied airborne animal- viruses, has been detected in air contaminated by infected- pigs and ruminants in both laboratory settings and farm environments. This single-stranded RNA (ssRNA) virus of the *Picornaviridae* family is excreted in all body fluids of infected animals and can become airborne directly from the animals or from the secondary-aerosolization of deposited-viruses or virus-laden particles. Other suspected sources of airborne -viruses, such as burning carcasses of infected animals , have not yet been identified formally as true sources because additional investigations are needed.

Poultry farms are also potential producers of virus-laden airborne-particles. The exotic Newcastle- disease virus (*Paramyxoviridae* family) was probably the first virus isolated from a naturally contaminated environment of poultry- houses sheltering infected birds. This 150-nm-diameter ssRNA virus was detected in air samples from 2 farms during an outbreak in Southern-California in 2002-2003. Air samples in and around broiler poultry-houses have also been studied for the presence of viruses such as *Escherichia-coli* bacteriophages, which are a fecal- contamination tracer. Other animals, such as bats (rabies- virus) , rabbits

(rabbit poxvirus), and mice (polyomavirus), have been studied as sources of bio-aerosols. These viruses can be released into the air directly from animals by their breathing, coughing, and sneezing or by secondary-aerosolization. It should be noted that the means of aerosolization has a critical impact on the aerodynamic- size and, thus, on the behavior of the airborne-particles. Given that virus-laden particles are a complex mixture of various components (salts, proteins, and other organic and inorganic- matter, including virus- particles), it is essential to realize that the size of the viral -particle itself does not rule the airborne-par-ticle-size.

Another study investigating pigs- infected with Aujeszky's disease-virus (*Herpesviridae* family; about 150 nm in diameter; double-stranded DNA [dsDNA] virus) found that the infectivity of the aerosols collected in each stage of the three-stage impinger varied over time. The investigators reported that the size- distributions of the aerosols in the three stages were comparable on day 2 of the infection but that there was an increase in infectivity associated with larger-particles on days 3 and 4. no clear association has been made between aerosol- infectivity and a particular size range. While single virus -particles exist in the air, they tend to aggregate rapidly. Aggregation speed depends on the size-distribution of the airborne particles, the concentration of the aerosol, and the thermo-dynamic conditions . infectious droplets exhaled by animals shrink rapidly with the lower-humidity outside the respiratory-airway, creating smaller aerosols. the size distribution of such naturally generated bio-aerosols depends on the sizes of the particles to which the microorganisms bind. This binding may occur by diffusion, impaction, interception, or electrostatic attraction.

Larger- particles may be relatively less hazardous than smaller ones. It has been shown on pig farms that a visually clean environment may be more contaminated by bio-aerosols than a visually dirty one " [1].

"To our knowledge, the oldest study on the sampling of airborne -viruses was performed with a laboratory setup using a chamber and an artificially produced aerosol of influenza- virus "[1].

MK Ijaz, et al., The survival of airborne human-corona-virus 229E (HCV/229E) was studied under different conditions of temperature (20 +/- 1 degree C and 6 +/- 1 degree C) and low (30 +/- 5%), medium (50 +/- 5%) or high (80 +/- 5%) relative- humidities (RH). At 20 +/- 1 degree C, aerosolized HCV/229E was found to survive best at 50% RH with a half-life of 67.33 +/- 8.24 h while at 30% RH the virus half-life was 26.76 +/- 6.21 h. At 50% RH nearly 20% infectious- virus was still detectable at 6 days. High RH at 20 +/- 1 degree C, it was found to be the least- favourable to the survival of aerosolized- virus and under these conditions the virus half-life was only about 3 h; no any virus could be detected after 24 h in aerosol. At 6 +/- 1 degree C, in either 50% or 30% RH- conditions, the survival of HCV/229E was significantly-

Table 1: Effects of RH relative humidity on infectivity of a selection of airborne viruses

Virus	Optimal RH for maximum infectivity	Family	Genetic material	Size (nm)	Envelope
Influenza virus	Low	<i>Orthomyxoviridae</i>	ssRNA (-)	80-120	Yes
Newcastle disease virus	Low	<i>Paramyxoviridae</i>	ssRNA (-)	150	Yes
Vesicular stomatitis virus	Low	<i>Rhabdoviridae</i>	ssRNA (-)	60 × 200	Yes
Japanese encephalitis virus	Low	<i>Flaviviridae</i>	ssRNA (+)	40-60	Yes
Porcine reproductive and respiratory syndrome virus	Low	<i>Arteriviridae</i>	ssRNA (+)	45-60	Yes
Semliki Forest virus	Low	<i>Togaviridae</i>	ssRNA (+)	70	Yes
Human corona-virus 229E	Mid-range	<i>Coronaviridae</i>	ssRNA (+)	120-160	Yes
Rotavirus	Mid-range	<i>Reoviridae</i>	dsRNA	100	No
Pseudorabies virus	Mid-range	<i>Herpesviridae</i>	dsDNA	200	Yes
Rhinovirus	High	<i>Picornaviridae</i>	ssRNA (+)	25-30	No
Poliovirus	High	<i>Picornaviridae</i>	ssRNA (+)	25-30	No
Picornavirus	High	<i>Picornaviridae</i>	ssRNA (+)	25-30	No
T3 coliphage	High	<i>Podoviridae</i>	dsDNA	60 (capsid)	No
Rhinotracheitis virus	High	<i>Herpesviridae</i>	dsDNA	200	Yes
St. Louis encephalitis virus	All	<i>Flaviviridae</i>	ssRNA (+)	40-60	Yes

enhanced, with the decay -pattern essentially similar to that seen at 20 +/- 1 degree C. At a low- temperature and high RH (80%), the survival pattern was completely reversed, with the HCV/229E half-life increasing to 86.01 +/- 5.28 h, nearly 30 times that found at 20 +/- 1 degree C and high RH. Although optimal survival at 6 degree C still occurred at 50% RH, the pronounced stabilizing effect of low -temperature on the survival of HCV/229E at high RH indicates that the role of the environment on the survival of viruses in air may be more complex and significant than previously thought [2].

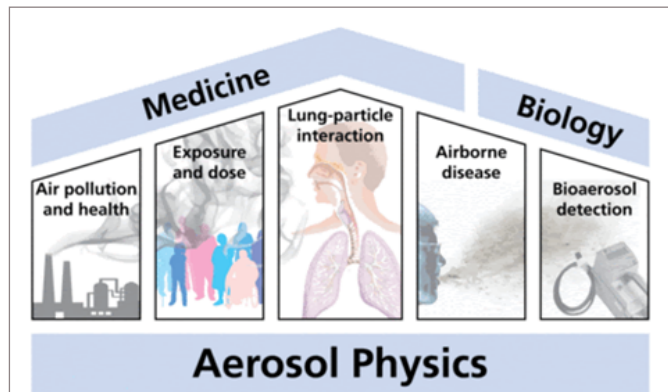


Figure 1: As showed in this figure aereosol physics cover different scientific discipline like Medicine, biology, chemistry and also physics.

It is not possible to study a respiratory virus disease without not consider also chemical physical properties in the interfaces of interaction virus – carriers.

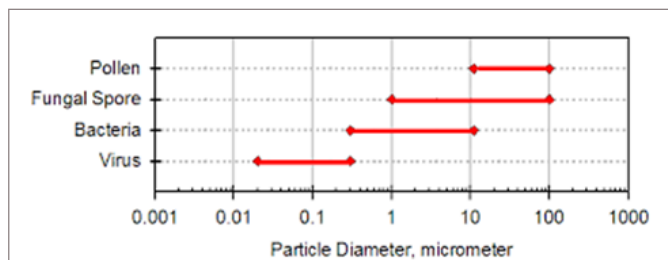


Figure 2: Related Bio-aerosols some references are of interest for the scope of this work: According E. Domingo: A primitive RNA world.

“A simplified -overview of a course of events that led to the origin of biological systems is depicted in Figure 3. The prebiotic synthesis of potential building blocks which might have been initiated earlier than about 5000 million years ago renders plausible the existence of a pre-RNA era that was then replaced by an RNA world in the late Hadean early Archean periods on Earth. This stage should have been followed by one in which RNA was complemented by DNA as a repository of genetic- information. Polymers other than DNA and RNA are also capable of encoding evolvable inheritable- information. Heterogeneous nucleic-acid molecules (including mixtures of ribo and deoxyribo-polymers) can give rise to functional nucleic acids. RNA-enzymes (ribozymes), such as RNA ligases can evolve from random sequence RNAs. The critical polymerization -reaction involves the formation of a phosphor-diester bond and release of pyro-phosphate—analogueous to the reactions catalyzed by the present-day RdRps and represent an incipient, primitive- anabolism. In support of a possible link between catalytic RNA-activities and solid mineral- surfaces in the origin of life, the catalytic activity of the hammerhead ribozyme of the Avocado Sun Blotch viroid was maintained when bound to the clay mineral montmorillonite.

Theories of the Origins of Viruses

Although not in a linear fashion, the number of nucleotides or base pairs in the genetic- material that presumably reflects the amount of genetic information relevant to confer phenotypic traits increased as evolution led to differentiated organisms. The major theories of the

origin of viruses are divided into 2 opposite categories: those that attribute virus origins to the early development of life, and those that propose that viruses arose when a cellular life was already in place. These 2 broad views that we may term “viruses without cells” and “viruses from cells” are not irreconcilable, although reconstruction of ancestral- developments is challenging. They can be divided into 5 main theories not all independent or mutually exclusive— which are summarized next.” [3].

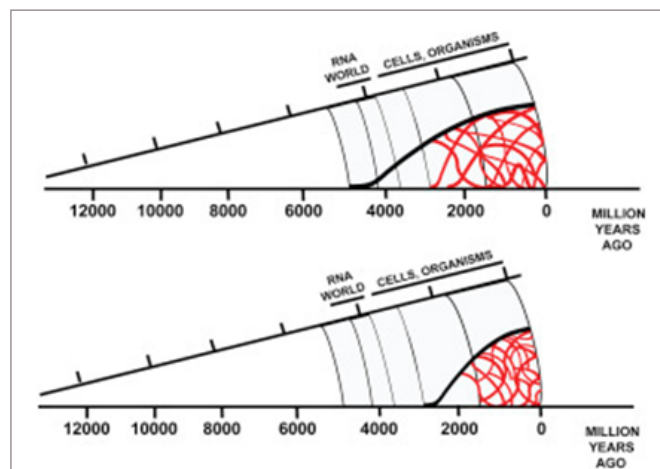


Figure 3: Two possible courses of events regarding when viruses first appeared and participated in the evolution of the biosphere.

The scheme of time frames and major biological events (RNA- world, first cells, and organisms) are those displayed in Figure 3. According to the upper- diagram, viruses (or previrus-like entities) arose together with the first (pre-cellular) replicating entities. According to the second diagram, viruses (or pre-virus-like entities) arose when a cellular life had already been established. Presence of virus is generically represented by the external, thick, black curves. The internal red, wavy lines represent generation, dominance, and extinction of multiple viral- lineages whose numbers and true dynamics will remain unknown. And in article Airborne Infectious Micro-organisms L.D. Stetzenbach, in Encyclopedia of Microbiology (Third Edition), 2009.

2. Bio-aerosols

“A bio-aerosol is an airborne collection of biological- material. Bio-aerosols can be comprised of bacterial cells and cellular fragments, fungal spores and fungal hyphae, viruses, and by-products of microbial metabolism. Pollen grains and other biological material can also be airborne as a bio-aerosol. Microbial aerosols are generated in outdoor and indoor environments as a result of a variety of natural and anthropogenic activities. Wind, rain and wave splash, spray irrigation, waste water treatment activity, cooling towers and air handling water spray systems, and agricultural processes such as harvesting and tilling are examples of activities that generate bio-aerosols outdoors. Indoors bio-aerosols are generated and dispersed mechanical and human activity. Industrial and manufacturing practices and bio-fermentation procedures can generate high concentrations of microbial -aerosols. Heating, ventilation, and air conditioning (HVAC) systems, water spray devices (showerheads and humidifiers), and cleaning (dusting, sweeping, vacuuming, and mopping) result in the transport of microbial -materials in the air. Talking and coughing generate bio-aerosols from individuals, some of which may be infectious. Facilities with medical, dental, or animal care practices can generate infectious microbial- aerosols.

The individual particle-size of particulate material in bio-aerosols is generally 0.3–100 μm in diameter; larger particles tend to settle rapidly and are not readily transported in the air. Virus particles are nanometer in size, bacterial cells are approximately 1 μm in diameter, and fungal spores are > 1 μm. These micro-organisms can be dispersed in the air as single -units, but are often present as aggregate -formations. The larger -aggregates have different aerodynamic properties than single-cell units; therefore their dispersal may be different than single-unit particles. Aggregates of biological- material also afford protection from environmental- stresses such as desiccation, and exposure to ultraviolet radiation ozone and other pollutants in

the atmosphere. Often bacterial -cells and virus particles are associated with skin cells, dust, and other organic or inorganic- material. During agricultural practices (during harvesting, and tilling), fungus spores are released from plant surfaces and the soil and raft on other particulate matter. This 'rafting' affects the aerodynamic characteristics and the survival of the cells in the bio-aerosol. When biological material is dispersed from water sources (splash, rainfall, or cooling towers and fountains), it is generally surrounded by a thin layer of water. This moisture layer also changes the aero-dynamic properties and aids in the survivability of the micro-organisms while airborne. Airborne- particulate will remain airborne until settling occurs or they are inhaled. Following inhalation, large airborne particles are lodged in the upper respiratory tract (nose and naso-pharynx). Particles <6 μm in diameter are transported to the lung where the 1–2 μm particles have the greatest retention in the alveoli.”

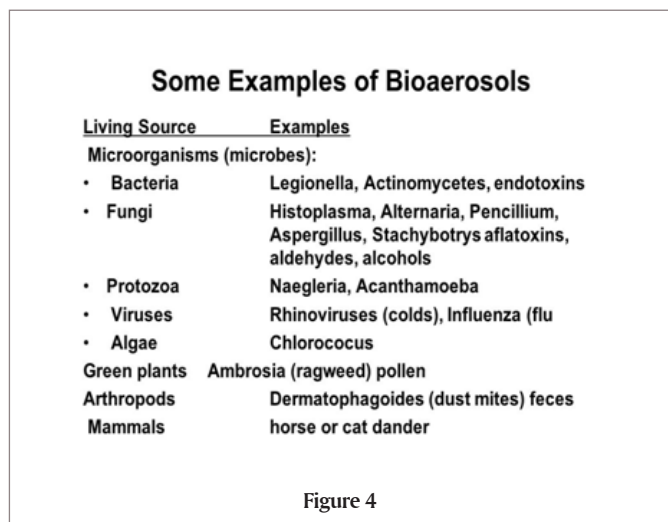


Figure 4

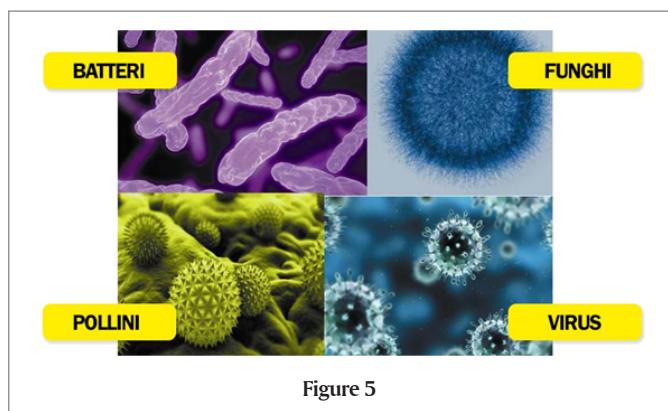


Figure 5

Prakriti Sharma Ghimire, et al. “Bio-aerosols such as airborne- bacteria, fungal spores, pollen, and others possess diverse characteristics and effects. A large gap exists in the scientific understanding of the overall physical-characteristics and measurement of bio-aerosols. Consequently, this review study aims to devise an appropriate approach to generate more scientific knowledge of bio-aerosols” [4].

Beijing [China]: Local authorities have said that the novel corona-virus can spread through direct transmission, contact- transmission, or aerosol transmission.

Shanghai officials reveal novel corona-virus transmission modes By Zhou Wenting in Shanghai | chinadaily.com.cn | Updated: 2020-02-08 “Confirmed transmission- routes of the novel-corona-virus include direct transmission, contact transmission and aerosol -transmission, a Shanghai official said on Saturday.

“Aerosol transmission refers to the mixing of the virus with droplets in the air to form aerosols, which causes infection after inhalation, according to medical experts,” said Zeng Qun, deputy- head of the Shanghai Civil Affairs Bureau.”

Bioaerosol Fundamentals:



Particle size and natural background concentration of bioaerosols:

Type of Bioaerosol	Size (μm)	Concentration (#/m ³)
Viruses	0.02-0.3	-----
Bacteria	0.3-10	0.5-1,000
Fungal Spores	0.5-30	0-10,000
Pollen	10-100	0-1,000

Source: Hinds, W.C., *Aerosol Technology: Properties, Behavior, and Measurement of Airborne Particles*, 2nd edition.

- Bioaerosol particles often occur as agglomerates, as clusters of organisms in droplets or attached to other airborne debris
- Bioaerosols can be subdivided into two groups:
 - viable: living organisms
 - nonviable: dead organism, pollen, animal dander, etc.

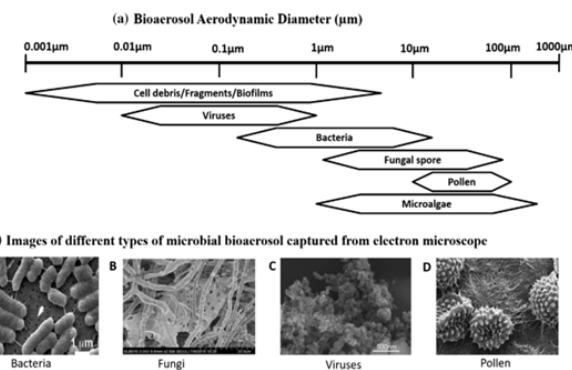


Figure 6

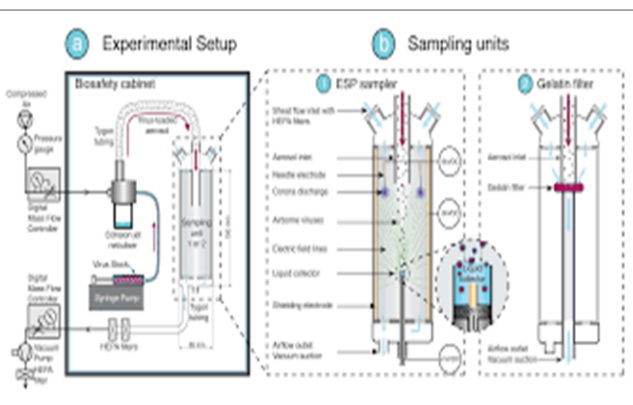


Figure 7: Schematics of the experimental setup. (a) Experiments are conducted in a bio-safety cabinet to keep all aerosolized- virus contained. The setup includes a Collison-nebulizer to aerosolize using compressed-air and viral solutions provided by a syringe pump. Tubing directs the airborne viruses into the sampling unit (b): (1) our ESP sampler or (2) gelatin- filters for comparison. A down-stream vacuum pump maintains a constant flow and under pressure inside the ESP sampler. (b) Details of the sampling units: (1) Our ESP sampler includes a three-electrode corona discharge electrostatic- precipitator to capture the aerosol particle directly into an integrated liquid collector with a miniaturized volume of 150 μL; (2) gelatin filters are used for comparative measurement of the total amount of virus effectively entering the ESP -sampler after nebulization. The filters are housed in a specific cassette placed at the aerosol -inlet. Ladhani L, Pardon G, Meeuws H, van Wesenbeeck L, Schmidt K, Stuyver L, et al. (2017) Sampling and detection of airborne influenza virus towards point-of-care applications.

According the new York times 5 oct 2020 Apoorva Mandavilli: 239 Experts With One Big Claim: The Corona-virus Is Airborne

“The WHO has resisted mounting evidence that viral- particles floating indoors are infectious, some scientists say. The agency maintains the research is still in-conclusive”.

It Is Time to Address Airborne Transmission of Corona-virus Disease 2019 (COVID-19)

Lidia Morawska, *et al.* We appeal to the medical-community and to the relevant national and international bodies to recognize the potential for airborne spread of corona-virus disease 2019 (COVID-19). There is significant potential for inhalation -exposure to viruses in microscopic- respiratory droplets (micro-droplets) at short to medium distances (up to several meters, or room scale), and we are advocating for the use of preventive -measures to mitigate this route of airborne- transmission. Studies by the signatories and other researcher- scientists have demonstrated beyond any reasonable doubt that viruses are released during exhalation, talking, and coughing in micro-droplets small enough to remain aloft in air and pose a risk of exposure at distances beyond 1–2 m from an infected- individual . at typical indoor-air velocities , a 5- μm droplet will travel tens of meters, much greater than the scale of a typical room, while settling from a height of 1.5 m to the floor. Several retrospective- studies work conducted after the severe acute respiratory syndrome corona-virus 1 (SARS-CoV-1) epidemic demonstrated that airborne- transmission was the most likely mechanism explaining the spatial- pattern of infections . Retrospective- analysis has shown the same for severe -acute respiratory syndrome corona-virus 2 (SARS-CoV-2) . In particular, a study in their review of records from a Chinese- restaurant observed no evidence of direct or indirect -contact between the 3 parties. In their review of video records from the restaurant, they observed no evidence of direct or indirect -contact between the 3 parties.

Many research studies conducted on the spread of other viruses, including respiratory- syncytial virus (RSV), Middle East Respiratory Syndrome Corona-virus (MERS-CoV) , and influenza , show that viable airborne- viruses can be exhaled and/or detected in the indoor- environment of infected patients . This poses the risk that people sharing such environments can potentially inhale these viruses, resulting in infection and disease.

There is every reason to expect that SARS-CoV-2 behaves similarly, and that transmission via airborne micro-droplets is an important pathway. Viral-RNA associated with droplets $<5\ \mu\text{m}$ has been detected in air , and the virus has been shown to maintain infectivity in droplets of this size. Other viruses have been shown to survive equally well, if not better, in aerosols compared to droplets on a surface . The current guidance from numerous international and national- bodies focuses on hand-washing, maintaining social- distancing, and droplet precautions. Most public health -organizations, including the WHO, do not recognize airborne- transmission except for aerosol-generating procedures performed in healthcare settings. Hand-washing and social- distancing are appropriate but, in our view, insufficient to provide protection from virus-carrying respiratory-micro-droplets released into the air by infected people. This problem is especially acute in indoor or enclosed- environments, particularly those that are crowded and have inadequate ventilation relative to the number of occupants and extended exposure periods. airborne -transmission appears to be the only plausible explanation for several super-spreading events investigated that occurred under such conditions , and others where recommended precautions related to direct -droplet transmissions were followed” [5].

3. Materials And Methods

Whit an observational point of view some relevant literature are analyzed and After producing an experimental hypotesys a global conclusion is submitted to the researcher. All literature comes from bio-medical databases (Pubmed and other opens journal) Some Preprint are included in reference due to the need to have rapid scientific data in actual second wave of Covid-19 pandemia.

4. Results

From literature:

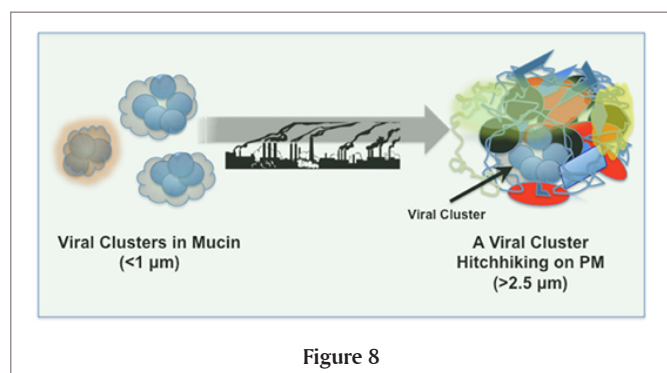


Figure 8

Airborne Particulate Matter and SARS-CoV-2 Partnership: Virus Hitchhiking, Stabilization and Immune Cell Targeting — A Hypothesis Z. Shadi Farhangrazi, Giulio Sancini, A. Christy Hunter and Seyed Moein Moghimi.

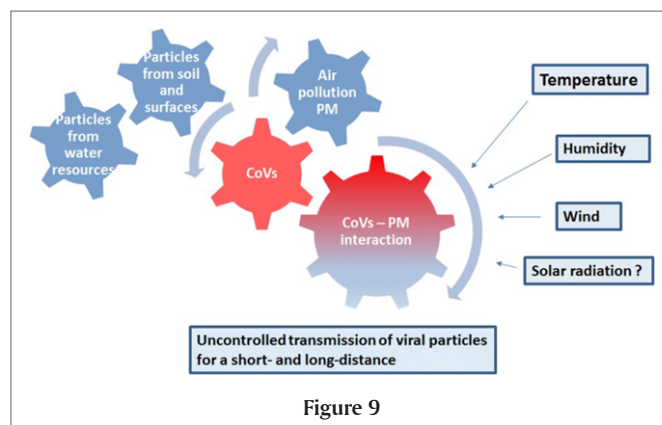


Figure 9

Tatiana Borisova, *et al.* interaction of SARS-CoV-2 envelope with PM is possible in water surrounding. After drying, PM can serve as a carrier for transmission of SARS-CoV-2 immobilized at their surface. PM and SARS-CoV-2 per se can enter human organism during nasal- inhalation, and they both use the same nose-to-brain delivery path-ways moving along axons directly to the brain, influencing the nervous system and exocytosis/endocytosis in nerve cells. Thus, PM can aggravate neurological symptoms of SARS-CoV-2 and vice versa, due to their identical nose-to-brain delivery mechanism and possible interference of neuronal effects. different types of PM because of their ability to interact with the plasma -membranes of nerve cells can facilitate unspecific SARS-CoV-2 entrance to the cells, and can influence envelope features of SARS-CoV-2. Detailed studies are required to analyze interaction of SARS-CoV-2 with PM [7]

Nguyen Thanh Tung, *et al.* Viable avian influenza viral RNA was found in PM up to 60 m downwind of commercial turkey farms using reverse-transcription (RT)-PCR and culture techniques. Influenza viral-RNA was detected in air samples collected approximately 2 km from the farms . A research study in the US reported that PM_{10} had higher estimated concentrations of avian influenza virus than $\text{PM}_{2.5}$, but $\text{PM}_{2.5}$ may be further aerielly transported. That work also reported transmission of the avian influenza virus via $\text{PM}_{2.5}$ within a state and between states . Viruses may be adsorbed through coagulation onto PM and remain airborne for hours or days, thereby increasing inhaled concentrations of virus via PM in the lungs. $\text{PM}_{2.5}$ may provide a good platform to “shade” and “carry” the SARS-CoV-2 during atmospheric- transport. PM containing SARS-CoV-2 could be a direct- transmission model in a highly polluted area [8].

Luigi Sanità di Toppi, *et al.* “One notable feature of all particulates PM is that they can convey (and release) toxic molecules and/or micro-organisms and/or spores and/or viral particles. These components can be absorbed or adsorbed by the particulate particles, depending on whether they enter them (where they are potentially solubilized), or whether they bind to the external surface. the particles can be broken, thus multiplying their polluting and carrying power. Particulate matter, especially fine/ultrafine/nano-particles, can enter the bronchi and the lung alveoli as well as the blood (both plasma and erythrocytes), the coronary- arteries, the heart, the lymphatic system, and, ultimately, almost all organs, with serious or very serious consequences for health (carcinogenic and/or teratogenic effects).

PM of various sizes can penetrate the respiratory tract, in some cases up to the pulmonary alveoli. The particulate matter of various sizes can penetrate the respiratory tract, in some cases up to the Pulmonary alveoli [9].

Nikolai Nikitin, *et al.* The human- infectious dose of the influenza- A virus, when administered by aerosol to subjects free of serum neutralizing antibodies, ranges between 1.95×10^3 and 3.0×10^3 viral particles. To determine the concentration of virus -particles in the air, the RT-PCR

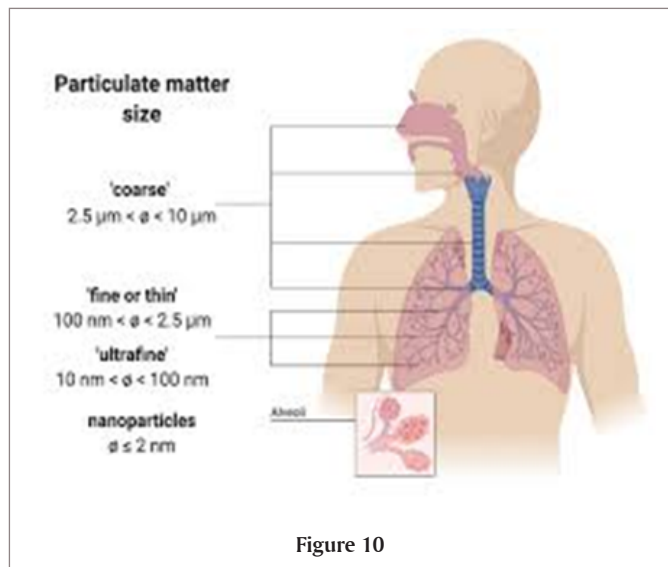


Figure 10

method is often used. However, RT-PCR analysis provides information on the total number of viral particles, but not on the number of infectious particles. Influenza virus genomic segments are chosen and packaged at random, whereby only parts of the virions are infectious. According to various scientific publications, data about the influence of the virus subtype on the effectiveness of influenza transmission are contradictory. The subtype-specific differences in influenza virus transmission were observed in animal models, and recipient animals did not exhibit a preexisting influenza virus specific immune response the pathogenicity of a virus subtype depends on the immune- status of the recipients (human). The second point is (when) how recently viruses of the same subtype circulated in the population previously.

In research studies conducted by Alford. *et al.*, volunteers were exposed to carefully titrated aerosolized influenza virus suspensions by inhaling through a face mask. The demonstration of infection in participants of the study was achieved by recovery of infectious viruses from throat swabs, taken daily, or by sero-conversion, that is, the development of neutralizing antibodies.

The use of carefully titrated viral- stocks enabled the determination of the minimal -infectious dose by aerosol inoculation. The approximate 50% human- infectious dose (HID₅₀-50% human infectious dose) of virus per volunteer was from 1 to 126 TCID₅₀ (the tissue culture 50% infectious dose). The dose for half of the volunteers was 5 TCID₅₀. The other half of the men, who had very low or non-detectable pre-inoculation antibody titers, were infected with 0.6 to 3 TCID₅₀. The study reliably shows that the human -infectious dose of the influenza- A virus, when administered by aerosol to subjects free of serum neutralizing antibodies, is approximately 3 TCID₅₀. The approaches used in this study allow the precise number of infectious- particles in the total number of particles to be determined. Ward, with co-workers, confirmed experimentally that three log₁₀ copies/mL corresponded to 1 TCID₅₀/mL. That is, one TCID₅₀/mL contains 1000 copies of the viral genome. According to other reports, the aerosol infection dose for humans was about 1.95 × 10³ viral genome copies, for approximately 300–650 copies of human-influenza viruses were contained in 1 TCID₅₀, according to previous studies . During the 2009-2010 influenza- season (from December to April), Yang, with coworkers, collected samples from a health centre, a day-care centre, and airplanes. The concentrations of airborne influenza viruses (A/PR/8/34 (H1N1) and A/swine/Minnesota/1145/2007 (H3N2)) were measured. The influenza A virus RNA was quantified by RT-PCR. Fifty percent of the samples collected contained the influenza A virus, with concentrations ranging from 5.8 × 10³ to 3.7 × 10⁴ genome copies per m³. The average concentration of the virus was 1.6 ± 0.9 × 10⁴ genome copies per m³, corresponding to 35.4 ± 21.0 TCID₅₀ per m³ air. According to Yang *et al.* , 1 TCID₅₀ of A/PR/8/34 (H1N1) stock was equivalent to 2.1 × 10³ genome- copies, and the ratio for the pandemic A/California/04/2009 (H1N1) strain was determined to be 452 ± 84 genome copies per TCID₅₀.

Using the measured airborne -virus concentration and an adult breathing- rate, Yang, with colleagues , estimated the inhalation doses during exposures of 1 h (e.g., the duration of a clinical visit), 8 h (a work-day), and 24 h to be 1.35 × 10⁴, 1.06 × 10⁵, and 3.2 × 10⁶ viral- particles (or 30 ± 18, 236 ± 140, and 708 ± 419 TCID₅₀), respectively. Compared with the aerosol HID₅₀ 0.6–3 TCID₅₀ , these doses are adequate to induce infection. In other words, over 1 h, the inhalation dose is estimated to be 30 ± 18 TCID₅₀ or about 16 000 particles of the influenza A -virus, which is more than enough to induce infection” (10)

Byung Uk Le, Corona-virus-Bio-aerosols Artificially- generated aerosols carrying corona-viruses have been studied with testing their stability. First, the Middle East -respiratory syndrome corona-virus (MERS-CoV) was aerosolized for 10 min and its viability was measured at 40% and 70% relative humidity (RH) conditions . The results revealed that MERS-CoV was stable at 40% RH. the virus viability was significantly lost at 70% RH. Second, SARS-CoV-2 was aerosolized for 3 hours and its viability was analyzed. It was found that the virus was viable even after 3 hours, with limited loss of viability. Corona virus genetic- materials in the air have been detected in several research studies. In a study by Azhar, *et al.*, in Saudi- Arabia, the MERS-CoV genome was detected in an air sample from a camel barn of an infected patient . In Wuhan (China) and Nebraska (US), SARS-CoV-2 nucleic acid tests conducted on air- samples gave positive results at an intensive care unit of a hospital in Wuhan (China) and in a patient room of a university medical center in Nebraska (US). In Florida (US), SARS-CoV-2 was detected in air samples at the Student Health Care Center at the University of Florida via RT-PCR analysis. In this study in Florida, the SARS-CoV-2 concentration was estimated to be 0.87 virus genomes/L air. in a study by Chia *et al.* in Singapore, SARS-CoV-2 was detected in air samples from the airborne- infection isolation rooms of infected patients via RT-PCR analysis and ORFlab assay. In a study by Liu (2020), SARS-CoV-2 RNA was detected in air samples from hospitals and public areas, such as department stores, in Wuhan (China) . The detection of corona virus genes in air samples implies that it is highly probable that corona-virus bio-aerosols were present at the sampling locations. In a study by Lednicky. *et al.*, the isolation of viable SARS-CoV-2 from air- samples of the surroundings (2 to 4.8 m away) of patients in a hospital was reported in Florida (US) [11].

Estimated minimum. size of particles (assuming homogenous distribution of viruses in released respiratory fluid particles and virus size of 0.09 μm) potentially carrying SARS-CoV-2 and corresponding aerosol suspension times.

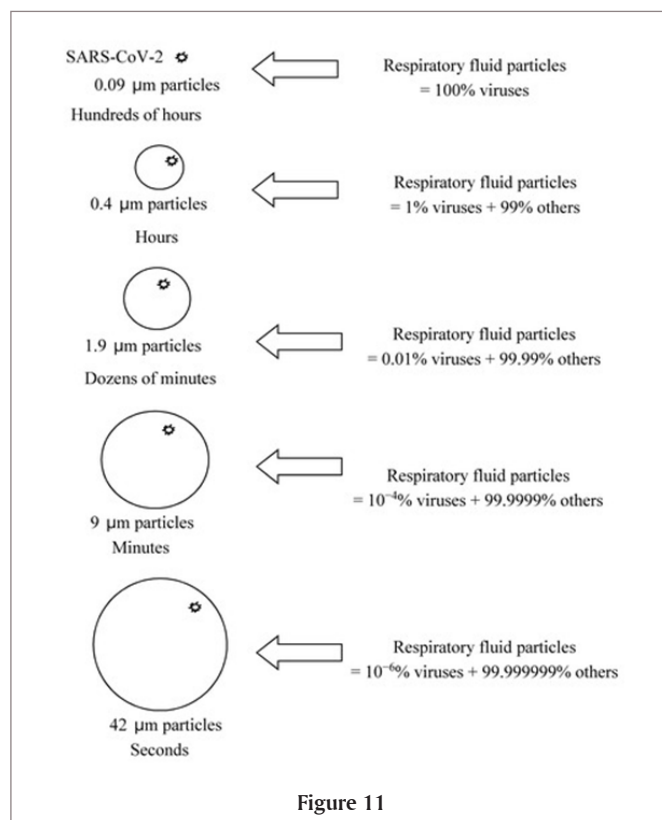


Figure 11

Michael Riediker, *et al.*, “The mean estimated viral load in micro-droplets emitted by simulated- individuals while breathing- regularly was 0.0000049 copies/cm³, with a range of 0.000000049 to 0.637 copies/cm³. The corresponding estimates for simulated coughing individuals were a mean of 0.277 copies/cm³ per cough, with a range of 0.000277 to 36 030 copies/cm³ per cough. The estimated concentrations in a room with an individual who was coughing- frequently were very high, with a maximum of 7.44 million copies/m³ from an individual who was a high emitter. regular breathing from an individual who was a high emitter was modeled to result in lower- room concentrations of up to 1248 copies/m³. In this modeling research study, breathing and coughing were estimated to release large numbers of viruses, ranging from thousands to millions of virus- copies per cubic meter in a room with an individual with COVID-19 with a high viral load, depending on ventilation and micro-droplet formation process. The estimated infectious risk posed by a person with typical viral load who breathes normally was low. The results suggest that only few people with very high viral load pose an infection risk in poorly ventilated closed -environments. These findings suggest that strict respiratory protection may be needed when there is a chance to be in the same small room with an individual, whether symptomatic or not, especially for a prolonged- period [12].

Marcelo I. Guzman, Various deposition mechanisms can exist, including inertial impaction, gravitational settling, Brownian motion, turbulent deposition, interception, and electrostatic attraction. The smallest particles (8 μm) are size dependently deposited from the nasal passage to the bronchioles. Multiple factors, (age, weight, sex, physical activity level, and disease state) impact respiration and deposition profiles . Larger- particles can be inhaled into the respiratory tract under exertion breathing because the oral cavity is larger and results in bypassing of the nasal- cavity filtration mechanism [13].

Silvia Comunian. *et al.*, The avian influenza -virus (H5N1) could be transported across long distances by fine dust during Asian- storms, and the correlation between PM concentration and the virus spread has been observed in the case of the spread of measles in China. PM2.5 concentrations in 21 Chinese -cities and the number of measles cases per day per city were studied. The analysis showed a positive correlation between those 2 factors. The 10 μg/m³ increase in PM2.5 per day is associated with a significant rise in the disease- incidence. A similar analysis of the children’s respiratory syncytial virus (RSV) spread in China in 2015 shows the same correlation. RSV is a virus that causes damage to the lungs and bronchitis. A positive -correlation between the virus and PM- concentration was observed. In fact, pollution increases the risk of RSV- infection.

A 2018 analysis, carried out in the Po Valley, associates hospitalizations and the number of new RSV cases with PM10 concentration. The data for the analysis were collected by ARPA (Regional Environmental Protection Agency) in the region. The results of this analysis showed that, in the designated period, the highest number of hospitalizations occurred in Milan, the city that had reached the maximum concentration of PM10. This study also shows a correlation between short and medium term PM10 exposures (in the two weeks preceding hospital admission) and increased risk of hospitalization owing to RSV- bronchiolitis among infants. There are several mechanisms by which PM induces an increase in infected cases. A mechanism can be that the virus is bound to particles and transported, if favored by climatic- conditions [14].

Tatiana Borisova. *et al.*, CoVs are large enveloped non-segmented positive-sense RNA viruses. Viral- envelopes consist of proteins and lipid components, and enveloped -viruses require the fusion of their lipid- envelope with the host cell membrane to entry the infected- cells interaction of SARS-CoV-2 envelope with PM is possible in water -surrounding. After drying, PM can serve as a carrier for transmission of SARS-CoV-2 immobilized at their surface. In this research study, we have suggested that lipid- constituents of the viral envelope can be very important for unspecific interaction of viral particles with different surfaces, including air -pollution particulate matter- (PM). It should be emphasized that this interaction capability can be inherent mainly to enveloped viruses. it was confirmed that air- pollution PM can travel across border for a long -distance and inhalation with fine and ultrafine- PM (the aerodynamic

diameter is less than 2.5 μm and 0.1 μm, respectively) is associated with many diseases, including neurological ones . The effect of fine dust -concentrations in the air in the Republic of Korea (2016–17) on the incidence of viral respiratory- infections caused by the human -corona-virus, respiratory syncytial virus, human metapneumovirus, adenovirus, rhinovirus, human bocavirus, human parainfluenza virus, and influenza-virus was investigated. It was concluded that when the weekly average concentration of fine dust increased, the incidence of infections by the human-corona-virus, human meta-pneumovirus, adenovirus, human bo-cavirus, human- parainfluenza virus, and influenza also increased . In the USA, the majority of the positive cases of highly pathogenic avian influenza (HPAI) H5N2 might have received airborne virus carried by fine air pollution PM, and these results provide insights into the risk of airborne transmission of HPAI virus via fine dust- particles.

In Beijing, China, association between daily PM2.5 (PM with size lesser than 2.5 μm) and influenza-like illness ILI risk was investigated using a generalized additive model. A strong positive- relationship between PM2.5 and ILI risk at the flu season was established, but the effect of PM2.5 differed across age groups [15].

Sima Asadi. *et al.*, These results show that dried influenza virus remains viable in the environment, on materials like paper tissues and on the bodies of living animals, long enough to be aerosolized on non-respiratory dust particles that can transmit infection through the air to new mammalian hosts [15].

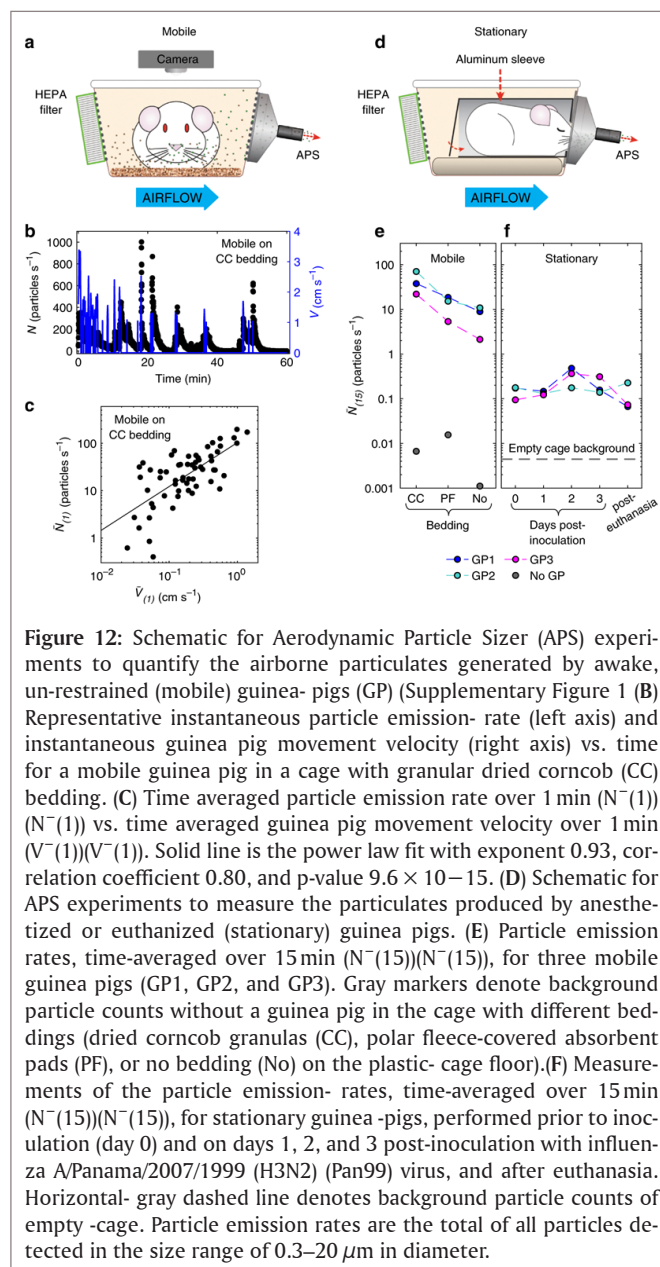


Figure 12: Schematic for Aerodynamic Particle Sizer (APS) experiments to quantify the airborne particulates generated by awake, un-restrained (mobile) guinea- pigs (GP) (Supplementary Figure 1 (B) Representative instantaneous particle emission- rate (left axis) and instantaneous guinea pig movement velocity (right axis) vs. time for a mobile guinea pig in a cage with granular dried corncob (CC) bedding. (C) Time averaged particle emission rate over 1 min ($N_{t}^{(1)}$) ($N_{t}^{(1)}$) vs. time averaged guinea pig movement velocity over 1 min ($V_{t}^{(1)}$)($V_{t}^{(1)}$). Solid line is the power law fit with exponent 0.93, correlation coefficient 0.80, and p-value 9.6×10^{-15} . (D) Schematic for APS experiments to measure the particulates produced by anesthetized or euthanized (stationary) guinea pigs. (E) Particle emission rates, time-averaged over 15 min ($N_{t}^{(15)}$)($N_{t}^{(15)}$), for three mobile guinea pigs (GP1, GP2, and GP3). Gray markers denote background particle counts without a guinea pig in the cage with different beddings (dried corncob granulas (CC), polar fleece-covered absorbent pads (PF), or no bedding (No) on the plastic- cage floor).(F) Measurements of the particle emission- rates, time-averaged over 15 min ($N_{t}^{(15)}$)($N_{t}^{(15)}$), for stationary guinea -pigs, performed prior to inoculation (day 0) and on days 1, 2, and 3 post-inoculation with influenza A/Panama/2007/1999 (H3N2) (Pan99) virus, and after euthanasia. Horizontal- gray dashed line denotes background particle counts of empty -cage. Particle emission rates are the total of all particles detected in the size range of 0.3–20 μm in diameter.

5. Science News

From research organizations Research exposes new vulnerability for SARS-CoV-2 Electrostatic interactions enhance the spike protein's bond to host cells August 11, 2020 Northwestern University.

The spike protein contains the virus' binding- site, which adheres to host cells and enables the virus to enter and infect the body. Using nanometer level simulations, the researchers discovered a positively-charged site (known as the polybasic cleavage site) located 10 nanometers from the actual binding site on the spike protein. The positively charged site allows strong bonding between the virus protein and the negatively charged human-cell receptors.

Leveraging this discovery, the researchers designed a negatively-charged molecule to bind to the positively- charged cleavage site. Blocking this site inhibits the virus from bonding to the host cell [17]

Sandhya Verma, *et al.*, The corona-virus-nucleocapsid (N) protein is a multifunctional viral- gene product that encapsidates the RNA genome and also plays some as yet not fully defined role in viral RNA replication and/or transcription. A number of conserved negatively charged amino-acids are located within domain III in the carboxy end of all corona virus N proteins. Previous studies suggested that the negatively charged residues are involved in virus assembly by mediating interaction between the membrane (M) protein carboxy tail and nucleocapsids Corona virus N proteins are phosphorylated. The proteins are highly basic, with isoelectric points (pI) of 10.3 to 10.7. A three domain structure for the protein has been proposed based on early sequence comparisons of MHV strains. The amino terminal and central domains of all corona virus N proteins exhibit an overall positive charge, whereas the carboxy terminal domain is acidic. Conservation of negatively charged amino acids within the carboxy ends of all corona virus N proteins suggests that the residues are functionally relevant. data from an earlier study suggest that the carboxyl end of the protein mediates interaction with the M protein during assembly, and the charged residues within the region were hypothesized to possibly facilitate the interaction. Within the carboxy-terminal 22 amino- acids of the MHV-CoV N protein there are eight negatively -charged residues [18].

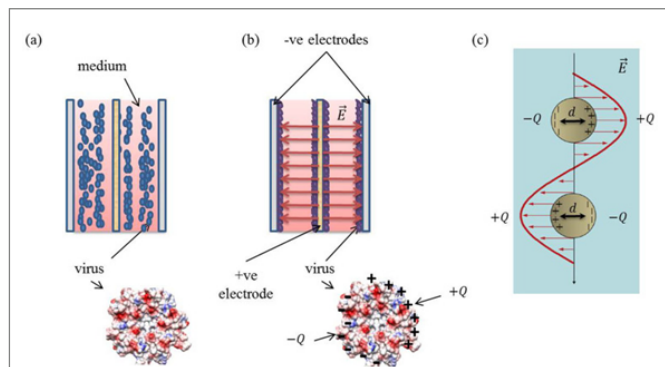


Figure 13: Illustration of the rationale of the electrical measurement for virus titer measurement and classification.

(a) Virions- distributions inside the coaxial-resonator in the absence of electric field. (b) Polarized-- virions when an electrostatic field is applied. A coaxial resonator has an inner conductor (the +ve electrode, where positive charges accumulate, +Q) surrounded by a hollow space that is surrounded by a conducting shield (-ve electrode, where the negative- charges accumulate, -Q). (c) Schematic of the polarized -virus particles inside an alternating current electric field. From Virus -detection and quantification using electrical -parameters.

Mahmoud Al Ahmad et al, Edris Joonaki, et al. Corona-virus- genomes are comparatively large for RNA viruses and severe acute respiratory syndrome corona-virus 2 (SARS-CoV-2) encodes an extensive complement of non-structural proteins (3-chymo-trypsin-like [3CL] protease, papain-like protease, etc.) as well as structural proteins as follows: spike (S) glycoprotein, envelope (E) glycoprotein, membrane (M) glycoprotein, and nucleocapsid (N) phosphoprotein.1 The SARS-CoV-2

spike (S) glycoprotein exhibits 76% amino -acid sequence identity with the SARSCoV S (Urbani strain) and 80% identity with S proteins of bat SARSr-CoV ZXC21 and ZC45.

1,2 CoV S-glycoproteins form club-shaped trimers and decorate the viral membrane, 3 giving corona virus virions their characteristic morphology. As a substantial component of the outer surface of the virion, S likely plays a critical role in adsorption of viruses onto the solid- surfaces under various environmental conditions.

For further clarification, Figure 14A depicts a central -slice through an electron micrograph of mouse hepatitis- virus, which exhibits the presence of S on the virion- surface. Viruses adsorb to surfaces through 2 main mechanisms, van der Waals (mainly mineral surfaces and, more importantly, electrostatic- interactions (charged surfaces in the presence of ions and or not neutral pH11–13). These 2 forces dictate the adsorption of viruses to surfaces. Although the interplay between these 2 forces is difficult to separate, indications of the interactions can be determined from past data. Viruses tend to be more hydro-phobic than proteins, thus they are attracted to metal- surfaces because of mainly van der Waals interactions as well as hydro-phobic effects. their ability to maintain the virus's viability and allow it to remain infectious is more of a function of the humidity and temperature, thus the surface energy of the water molecules plays a large- role in the interaction between a virus- particle and a surface.

SARS-CoV-2 virions can be adsorbed onto metal- surfaces (gold and stainless -steel) in addition to hydroxyl functional group- and oxygen-containing substrates (wood, cotton, paper, and glass) depending on the surface chemistry and environmental -conditions (bulk fluid pH, surface charge, temperature, etc).Hydrogen bonding plays a key role in the adsorption of viruses to the hydroxyl-containing surfaces and in the presence of an aqueous phase thin film- layer. The strength of the bond to the surface would be high in the presence of -O-H - -O bonding, particularly in pH environments where the carboxylic acid on the virus is de-protonated (typically above a pH of 4). At neutral pH, most viral particles have a net negative charge because they have an isoelectric point below 7. However, due to the large- size of virus particles and their large variety of surface- proteins, there are still multiple patches of positive and negative charge in the pH range where viruses are stable (typically from pH 5–8). NH_2 , NH_3^+ , COOH , and COO^- groups of amino acids in the SARS-CoV2 S-protein drive adsorption onto the solid surfaces through double electrostatic interactions between the virion's ionized surface-active species and the oppositely charged surfaces, as well as hydrogen bonding based on the surface characteristics. at neutral pH values, the negatively -charged virus particles would be adsorbed significantly less on a stainless-steel surface because of electrostatic-repulsion, given that both virion and substrate surface have negative charges. With augmentation of the cation concentration, however, the repulsion would be decreased, and the quantity of adsorbed viruses would increase. Figure 14 presents the potential molecular- interactions between the SARS-CoV-2 viral proteins and solid surfaces at different pH values and fluid chemistries. As denoted in Figure 14 A, at pH values below the isoelectric- point, the overall charge of SARS-CoV-2 could be positive, given that both the carboxylate and amine groups on the outer surface are protonated, and hydrogen bonding would be formed to hydroxyl-containing surfaces such as wood, cotton, or paper.

At pH environments above the isoelectric- point , the outer surface of virions would be deprotonated and therefore negatively- charged and cannot be adsorbed on the surface with the same charge. Accordingly, lower virus -adsorption onto the surfaces would occur at higher pH values.

Instead, they can interact strongly with divalent and/or monovalent cations if they existed in a brine electrolyte solution (more details are presented in the following section). The charge and counterions from the electrolyte could lead to thinner double layer and lower repulsion forces, and again hydrogen bonding formed to surface hydroxyl groups, which results in promoting the virus- adsorption process. The gold surface of an electrode in the quartz crystal microbalance (QCM) biosensor, which works on the basis of the oscillating frequency alteration could be employed for monitoring of the virus -surface adsorption and desorption phenomena with or without the presence of negatively-charged surface active species in the liquid- phase [20].

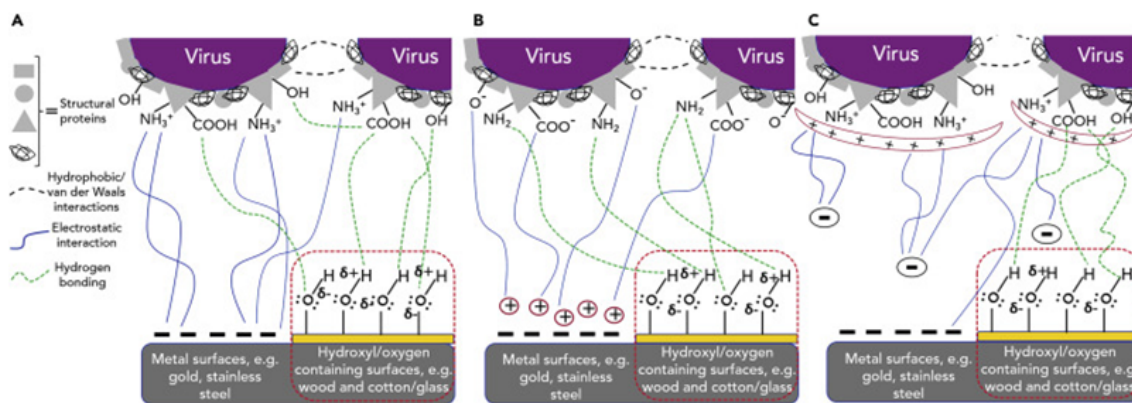


Figure 14: Molecular- Interactions at SARS-CoV-2 Viral Interfaces in Different Environmental Conditions- Model of the potential molecular interactions among viruses and between virus and different solid- surfaces having negative -surface charge and/or hydroxyl functional groups at (A) relatively low pH environment, below the isoelectric -point; (B) relatively high pH- condition, above isoelectric point in presence of external ions (salts); and (C) way below the isoelectric point in the presence of potential chemistries (for removal from surface purposes) with negative-surface charge.

Energy landscape theory of SARS-CoV-2 complexes with Particulate Matter. Gianluigi Zangari del Balzo This manuscript was compiled on June 24, 2020 Preprint ResearchGate

“The presence of multiple non-covalent interactions between SARS-CoV-2 and PMs is a source of cooperativity between them. Only when these interactions cooperate is a stable 235 single conformation produced, that of the complex [SARS-CoV- 2]⊕[PM]. The complex created is therefore much stronger than might be expected from the sum of their individual- strengths. This can explain the rapid spread of the pandemic in the areas of the greatest- pollution. This exceptional cooperative optimization can also explain the severity and difficulty of treating the forms of interstitial pneumonia that occur in Italy and worldwide. But not only that, it could perhaps also help us understand the origin and initial mutations of SARS-CoV-2” [22].

6. Experimental Project Hypotesis

In order to verify the effect of electrical charge on virus surface and the entity of ligand force between virus and carriers (in airborne condition) it is possible to think to 2 closed environment with virus in aerosols or with carrier like particulate matter:

- 1) environment under standard condition
- 2) environment uder with modified condition (differente level low-, moderate,high): Electrical charge influence, wind flux Chemical phisic properties (pressure temperature) humidity and other that can be applied.

After significative time exposure this aerosols must to be tested to verify the PM particle viral binding whit specific nalitical method. The PM must to be separated from the aereosol according their SIZE. A Different data in the 2 enviroment show the influence on weak links on virus interaction with PM.

7. Discussion

It is interesting that in airborne transmission the smaller particles are more dangerous then the larger ones: The smaller particles can introduce better in deep lungs zones since alveoli . See behavior of PM 2,5 vs PM 10. Chemical physical properties of virus surface are involved in the link – binding whit carriers like PM. This link is also responsible in the severity of disease. The composition and properties of virus surface are directly linked with airborne Pattern. WHO officially recognized direct contact and by droplet primary way of corona-virus transmission while airborne less involved. But related the preventive measure adopte the great number of death worldwide are clearly explained?

8 . Conclusion

Some parametere are relevant in evaluating airborne profile of some respiratory virus transmission. Many parameters are involved but also need to be deeply studied are: Virus size, envelope properties , electrical charge, chemical -physic interaction between virus and carrieres, attractive forces and repulsion, weak binds this properties must to be considered in adoption of preventive measure. Not only medicine and biology but also chemistry and phisic science can help in better explain some characteristic of respiratory virus, the law that regulate their diffusion and permanence time in aereols or linked with carrier. Chemical composition of virus envelope, kind of chemical links with carriers, Electrical charge, repulsion forces , and environmental factors influence airborne characteristic of some respiratory virus as well as time of permanence in aereols. H- bindings, van der waals interations, hydrophobic , electrostatic interactions are common weak link between virus particles and carriers. It is to conclude that is not possible control a corona-virus pandemia without not consider this last factors. The same severity of disease is related to the chemical -phisical link between virus and PM and the therapeutic strategy also.

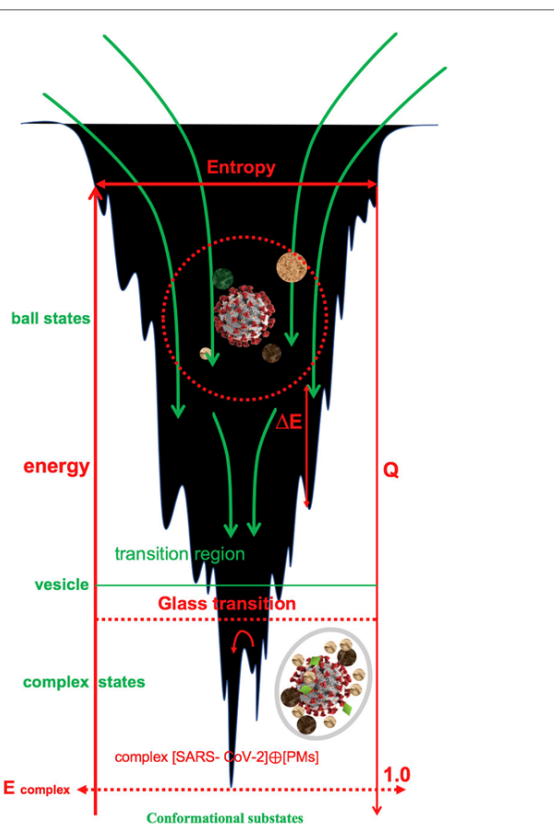


Figure 15: Energy rugged funnel landscape for a realistic [SARS-CoV-2]⊕[PMs] complex formation.

Conflict of Interests

No

Ethical Consideration

This work is produced under all international ethical rules, that can be applied.

Clarification

This work is produced without any diagnostic or therapeutic intent only to produce research hypotheses to be submitted to other researchers.

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