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The Concept of a Compound Autopsy Experimental Laboratory for SARS-CoV-2 Aerosolization Studies

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Abstract

This viewpoint material suggests the concept of a compound autopsy experimental laboratory for SARS-CoV-2 aerosolization studies. The autopsy room can provide at least three experimental models: wide open mucosa during the autopsy procedure, an experimental platform of sealed at the larynx level of the body's upper respiratory tract, and a frozen section cryostat chamber, as an additional experimental site for an emulation the aerosolization process. The autopsy room can be merged with a virology laboratory. The goal of the compound autopsy experimental laboratory could be to study the conditions for SARS-CoV-2 aerosolization. A positive result could lead to a variety of additional studies on SARS-CoV-2, as such the viability of an aerosolized virus. The epidemiological implications of these studies are difficult to overestimate. The method of such study would provide reliable reproduceable experimental data for use now and in a variety of applications in the future.

Keywords

SARS-Cov-2, Aerosolization, Autopsy, Experimental Platform

1. Introduction

SARS-CoV-2 aerosolization still remains at the level of the hypotheses [1]. Voices that try to connect the COVID-19 pandemic spread to the transmission route of aerosols are becoming increasingly louder [2]. At the same time in a different review article, professionals in aerosols studies underline that so many unanswered questions remain [3]. The words "might" and "may" are dominating clinical and epidemiological observations. Just some quotes: "Here we would like to pose a simple question: what role do aerosols play in transmission of COVID-19?" Or "… if pre- or asymptomatic infected individuals do not sneeze or cough, how do they generate aerosols?" One of the questions stands out: "What animal models are best for simulating airborne human transmission of COVID-19?"

What about looking at this unfortunate situation of the pandemic from a different perspective? Anatomic pathology, medicine, epidemiology, and human society need reliable scientifically supported data about SARS-CoV-2 virus aerosolization. It is necessary to develop a method that traces how the SARS-CoV-2 virus incorporated in infected person's mucosa becomes airborne. Certainly, it is very complex, if not impossible to do such a study with a sick patient, but neither an experimental animal nor tissue culture can completely substitute a human body. The autopsy of a deceased from COVID-19 can be as an experimental platform for such a study.

According to the literature data obtained regarding autopsy safety during the pandemic in the first half of 2020, recommendations were almost universal for postmortem performance on autopsy tables/workstations with a re-

verse-flow air handling system in negative-pressure ventilation rooms [4, 5, 6]. Only aerosolization can be considered as a source of SARS-Cov-2 virus contraction during an autopsy. Apparently, the prevention of an airborne infection is in the background of these recommendations.

In the tranquil situation of an autopsy room, some sophisticated experiments can be carried out to obtain significant information about the aerosolization of the virus. Figure 1 presents the main directions of studies on the autopsy experimental platform depending on positive or negative result of initial studies.

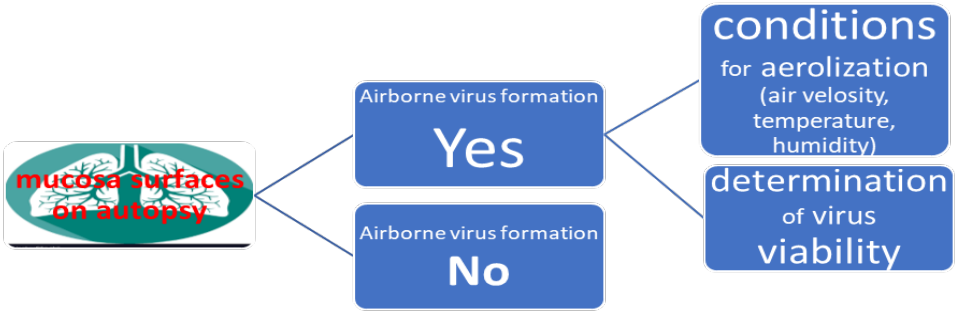

Figure 1. The main directions of studies on the autopsy experimental platform.

A YES result would shed light on the possibility of aerosolization in principle. Manipulations with air flow above large simultaneously opened mucosa surfaces with specially designed methods of virus capturing would provide the answer to the main question of the virus aerosolization "take off" during an autopsy. A NO result would diminish the fear of infection inoculation for workers and retract administrative limitations in the performance of autopsies.

2. Compound Autopsy Experimental Laboratory Concept

Below is the concept of a compound autopsy experimental laboratory structure and the possible fields of application. The autopsy experimental model would sound strange to people uninvolved in postmortem practice and disrespectful to the deceased. However, having participated in thousands of autopsies as a pathologist, the postmortem procedure for this author means a regular work event.

After accomplishing the medical or forensic goals of an autopsy, the body of the deceased becomes the object of scientific various specific fields of study, thus merging for example with a virology laboratory. Apparently, medical ethics and legal non-maleficence principles must be taken into account. After the experimental studies are over, the body is closed for appropriate funeral arrangements.

Three components form the basic structure of the autopsy experimental platform: an autopsy table in the morgue, a laboratory's biosafety cabinet for virology studies, and engineering controls for managing experiments with air circulation speed, room temperature, and humidity. The engineering controls of an autopsy room can provide the necessary conditions for creating manipulations to imitate the clinical "aerosol generating procedure" in a hospital ward or the spreading of viruses by a sick or asymptomatic individual.

As a new direction in investigative pathology, such a compound laboratory requires specific substrates for virus cultivation. Depending on the concrete experimental study, other instruments would need to be included, such as sensitive particle registration devices. It is not necessary for all components of the compound autopsy virology laboratory to be located in the same room as aerosol-capturing devices. The rest of the equipment can be in adjacent rooms or even further away if the technology allows.

Due to the instigation of the virus aerosolization, the biosafety measures should be almost at the BL-3 level, along with all rules of the negative pressure ventilation and air filtration. This article suggests only the principle of such an experimental platform. Details of the virology laboratory organization and design should be worked out during the implementation under specific conditions of the institution.

The initial SARS-CoV-2 aerosolization studies can be three main areas of compound autopsy experimental laboratory application: wide open mucosa surfaces; nose, nasopharynx, glottis, and larynx; and frozen section cryostat chamber (Figure 2). There could be different areas of application depending on the goal of studies.

3. Wide open mucosa platform

The autopsy dissection provides a unique opportunity to simultaneously expose a wide-open mucosa surface from the trachea to the bronchiole supposedly containing SARS-CoV-2. By applying variations to experimental

changes in the air velocity, the study could obtain data on the discharge of the virus from the mucosa surfaces to imitate spreading, especially a super-spreader.

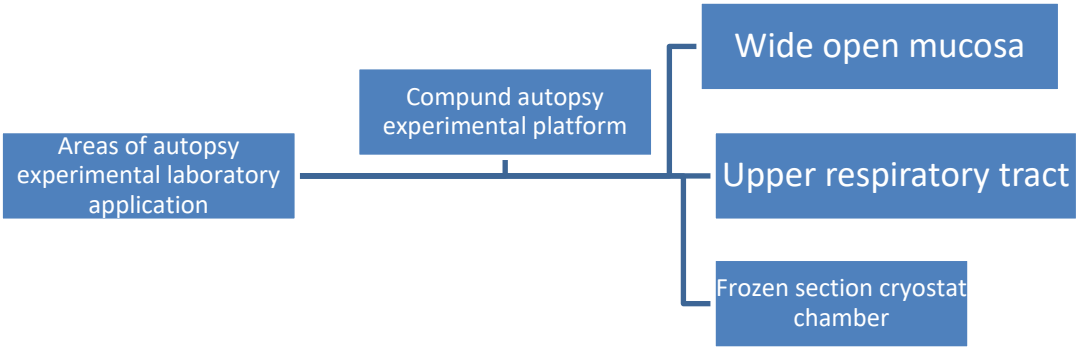

Figure 2. The main areas of compound autopsy experimental laboratory application.

Research on infectious disease aerosols already has some tools that can be used to collect airborne viruses (transmissible gastroenteritis and porcine coronaviruses) from heating, ventilating, and air conditioning (HVAC) filters in experimental apparatus and to sample these filters using AGI-30 All-glass and BioSampler impingers as well as frit bubblers [7, 8]. These have been examined using a Collison nebulizer or a plastic medical nebulizer. However, it would be impossible to employ a nebulizer on the autopsy experimental platform because, apparently, SARS-CoV-2 studies would require more sensitive samplers. For example, employing an investigation model of droplet bioaerosols in stabilized droplet clusters with a Axio Zoom V16 fluorescence microscope [9] or laser light-scattering experiments [10]. Those are only examples of an experimental platforms without specification of the details of the methodology that would be superfluous in the presentation of the concept. There might be different methodological approaches to conduct the experiments.

4. Upper respiratory tract's experimental platform

For understandable reasons, the upper respiratory tract is the most important and data-rewarding area of experimental studies examining SARS-CoV-2 aerosolization. The goal of the head/neck platform is to imitate as much as possible the real clinical and epidemiologically significant situation to study the discharge of virus containing droplet nuclei from the upper respiratory tract in the person's exhale. Although the larynx is usually a part of the evisceration complex, in the case of an experimental model, the larynx should be excluded because it does not have substantial significance in the final postmortem diagnosis of COVID-19 deceased.

Left in the cadaver's body after the regular evisceration procedure, the entire head/upper respiratory tract part is sealed at the level of the lower larynx (cricoid cartilage) or the upper trachea's cartilage rings. During experiments, an air composition close by components to the human's exhaled gas (78% nitrogen, 16 % oxygen, 4% percent carbon dioxide) is propelled through a tube inserted into the sealed head/neck complex (Figure 3). In this variant of the experimental autopsy platform, the head/neck part of the body would be like an aerosol spray nebulizer can. When the nasopharyngeal area is examined, the mouth is closed.

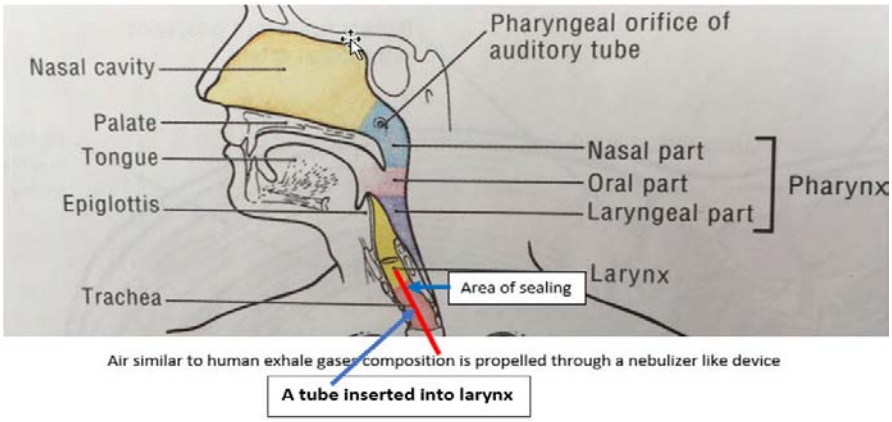

Figure 3. The upper respiratory tract's experimental platform.

There can be numerous variations in velocity, temperature, and humidity. The collected and examined exhaled discharge can present data of SARS-CoV-2 aerosolization particles. The samples are examined using aerosols and virology science methodology. The sample collection would be different from the wide open mucosa platform due to opportunities of proximity to the area of discharge of aerosol particles.

Attempts to develop a methodology similar to the presented model already exist. For example, a study was conducted to examine the effectiveness of face masks in obstructing respiratory jets [11]. The pressure impulse was emulated a cough or a sneeze to a hollow manikin head with a circular opening representing the mouth. The manikin was padded on the inside to approximate the internal shape and volume of the nasal and buccal cavities. The emulation was delivered via a manual pump or other sources, such as an air compressor or pressurized air canister. A cough simulator has also been developed for studies on the efficacy of face shields against cough aerosol droplets, which allows researchers to test ventilation and disinfection issues [12].

5. Frozen section cryostat chamber platform

The set of experiments that can be conducted in the closed cryostat chamber is enormous due to the variety of options available, starting with changes in air flow velocity using remotely managed ventilators. The cryostat is designed for changes in temperature in the chamber. Sectioning creates different temperature conditions through friction. The yes/no result of SARS-CoV-2 aerosolization would have useful consequences for regular practice in surgical pathology and other laboratories where cryosection is used on a daily basis.

Apparently, the sample's collection instruments will be different from the standard aerosols' science practice due to the limited size of the chamber. Their minimization and different sensitivity of aerosol sampling registration devices open a window for innovations that might find use beyond this laboratory.

6. Instead of Discussion and Conclusion

There would be definite safety issues, which would require BL-3 laboratory set of PPE and the laboratory's room design. This laboratory itself should be separate from the autopsy room itself.

These examples are presented that it is time to merge the experience of environment scientist's studies with actual SARSR-CoV-2 aerolization problem. This is a blue print of the experiment, which requires significant methodology work. Of course, it is easier to project in a hypothesis than carry out such experiment.

The crucial questions relating to safety and epidemiology are the direct mechanism for transforming a virion into airborne droplet nuclei. The SARS-CoV-2 virus is not sand on a beach. It is incorporated into the mucinous content of the predominantly respiratory tract's epithelial surfaces. The SARS-CoV-2 virus is in the mucus "envelope", which is made up of 95% water, although mucus contains other ingredients. The quotation marks are used here to prevent confusion with the viable virus's protein-lipid envelope. In water content, molecules follow the kinetic energy's Brown-like motion when the lightest and fastest can overcome Van der Walls's power to leave through the evaporation of mucus as < 5-10 μm water droplets.

This physical picture is far from a real biological situation. Even erythrocytes, the simple denucleated cell in the bloodstream, flow in aggregates; they look separate only on the glass slide. Before leaving the epithelial surface, the potential droplet nuclei are in adhesiogenic relationships in the mucus. The intimate mechanism of the process that prompts the virus to leave its comfortable dwelling at the "mucinous airport ground" for uncertain floating in the air is still awaiting serious experimental science. Without understanding these relationships, many epidemiological, diagnostic, and even treatment issues cannot be solved. The main epidemiology problem is the asymptomatic super spreaders.

No autopsy should be wasted as a source of additional knowledge about the COVID-19 disease. The quote MortuiVivos Docent ("the dead teach the living" from Latin), which is written on the entrance to the autopsy room in the Mount Sinai Hospital in Chicago, where I worked, would sound "the dead help the living" to fight the epidemics. A compound autopsy experimental platform could be the much needed step ahead in receiving reproducible scientific data on the current SARS-CoV-2 aerosolization problem. This method could contribute to studies of future epidemics.

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