# HANDLING OF SPECIMENS FROM COVID-19 SUSPECTED OR INFECTED PATIENTS

**Purpose of the document**
The purpose of this document is to provide guidance through procedures in handling COVID-19 infected and suspected samples, as described elsewhere (Cossarizza et al., 2020)

**Background**
SARS-CoV-2 belongs to the Coronaviridae family and is taxonomically related to the subgenus Sarbecovirus (Khan et al., 2020). This is an enveloped virus containing a single-stranded positive sense RNA as viral genome. Virions are spherical, with the spiked glycoprotein embedded in the envelope. Additional viral proteins include envelope, matrix, and nucleocapsid. The presence of SARS-CoV-2 RNA has been demonstrated by RT-PCR in bronchoalveolar lavage fluid, fibrobronchoscope brush biopsy, sputum, nasal swabs, pharyngeal swabs, feces, blood. Infection occurs primarily via direct contact or through droplets spread by coughing or sneezing from infected individuals. Importantly, viral load in nasal and throat swabs from an asymptomatic patient is similar to that of symptomatic patients, indicating that infected persons with no symptoms can transmit the virus.

**Stability of SARS-CoV-2 in aerosols and on various surfaces**
It has been reported that the virus can remain viable and infectious in aerosols for hours, and on surfaces up to days (depending on the inoculum shed) (van Doremalen et al., 2020). Human coronaviruses in general are known to persist on inanimate surfaces such as metal, glass or plastic for up to 9 days.

**Risk Assessment**
Each laboratory should conduct a local (i.e. institutional) risk assessment to ensure it is competent to safely perform the intended testing with appropriate risk control measures in place.

**Laboratory biosafety**
All laboratory personnel should adhere to appropriate biosafety practices. National guidelines on laboratory biosafety should be followed under all circumstances. For general information on laboratory biosafety guidelines, refer to the WHO Laboratory Biosafety Manual, 3rd edition.
When handling and processing specimens, including blood for serological testing, laboratory practices and procedures that are basic to good microbiological practices and procedures (GMPP) should be followed.

Non-propagative diagnostic laboratory work including sequencing, nucleic acid amplification test **(**NAAT) on clinical specimens from patients who are suspected or confirmed to be infected with Covid-19 should be conducted in a Biosafety Level (BSL)-2 laboratory. Initial processing (before inactivation) of all specimens including those for sequencing and NAAT should take place in a validated Class II biological safety cabinet (BSC) within a BSL-2 laboratory.

Handling of material with high concentrations of live virus (such as when performing virus propagation, virus isolation or neutralization assays) or large volumes of infectious materials should be performed only by properly trained and competent personnel in a containment laboratory with inward directional airflow, i.e. BSL-3.
**Cell sorting of all samples containing known aerosol pathogens must be performed in a BSL-3 laboratory.**
**Flow cytometry acquisition of any non-fixed cells from patients suspected or confirmed to be infected with Covid-19 must be handled as for sorting in a BSL-3 laboratory.**

**Personnel protection**
- All Laboratory personnel must wear **personal protective equipment (PPE)**.
- When working in the laboratory area it is mandatory to wear disposable gloves, eye protection, disposable laboratory coat, and surgical mask, to prevent the spread of unwanted droplets.
- During specimen manipulation in a Class II BSC it is mandatory to wear two pairs of disposable gloves, disposable laboratory coat, surgical mask, and eye protection.

- At the end of the procedure in a BSC, the external layer of gloves is removed and discarded into the waste located inside the BSC.
- Before leaving the laboratory, protective clothing must be removed and the personnel must wash hands thoroughly with liquid soap and running water for at least 20 sec.

**A potential exposure to infectious materials, or any sort of accident has to be immediately reported to the head of the laboratory for the appropriate evaluation.**

**Laboratory Working Areas**
- A distance of at least 2 m has to be maintained between people inside the lab and, if possible, the presence in each room should be limited to one person only.
- Designated working zones have to be respected.
- Eating, drinking, smoking, handling of contact lens, applying cosmetics, playing with the phone or chatting on social networks are absolutely prohibited.

**Cleaning and decontamination of working areas**
- Working surfaces must be accurately cleaned with 0.5% bleach (1/10 volume dilution of household bleach in tap water made fresh daily) for 10 min and then with 70% ethanol for 2 min when work is finished.
- Liquid waste must be decontaminated with 0.5% bleach for 20 min.

**Spills decontamination**
- Spills must be decontaminated with 1% bleach (1/5 volume dilution of household bleach).
- Apply paper towels to the area and soak the area with disinfectant (1% bleach), starting at the perimeter and working towards the centre. Allow 30 minutes of contact time before continuing to clean with additional applications of disinfectant.
- Dispose of all contaminated materials into biohazard trash.
- Clean with 70% ethanol (allow complete drying before disposal).

**Handling and processing of specimens from patients with suspected or confirmed Covid-19**
- Specimens are defined as, but not limited to, blood, serum, plasma, tissues, feces, urine, sputum, mucosal swabs or washes/secretions.
- Handling of any unfixed samples (specimens) from patients with suspected or confirmed COVID-19 must be done in a certified Class II BSC within a BSL-2 laboratory.
- All procedures with a high likelihood to generate aerosols or droplets, e.g., vortexing, mixing, sonication must be performed in a certified Class II BSC.
- Centrifuge buckets must be sealed for centrifugation, and specimens are centrifuged in securely capped polypropylene tubes that are loaded and unloaded in a Class II BSC.
- The BSC is daily equipped with an internal waste containing 0.5% bleach (1/10 volume dilution of household bleach in tap water made fresh daily) where any possible contaminated biological material has to be discarded.
- Only disposable plastic ware and pipettes may be used, which are decontaminated into the waste located inside the BSC.
- On completion of work in BSC, the internal waste is closed and discarded into a biosafety waste container.
- Surfaces are decontaminated with 0.5% bleach (1/10 volume dilution of household bleach n tap water) for 10 min and then with 70% ethanol for 2 min.

**Packaging and Transport**
- Patient specimens from suspected or confirmed cases must be transported to and between laboratories as UN3373, “Biological Substance, category B," and must be placed in two secondary containers to minimize the potential for breakage.
- Opening of containers is performed inside a certified Class II BSC.

**References**


Cossarizza, A., Gibellini, L., De Biasi, S., Lo Tartaro, D., Mattioli, M., Paolini, A., … Mussini, C. (2020). Handling and Processing of Blood Specimens from Patients with COVID-19 for Safe Studies on Cell Phenotype and Cytokine Storm. *Cytometry Part A*, (3). https://doi.org/10.1002/cyto.a.24009

Khan, S., Siddique, R., Shereen, M. A., Ali, A., Liu, J., Bai, Q., … Xue, M. (2020). Emergence of a novel coronavirus, severe acute respiratory syndrome coronavirus 2: Biology and therapeutic options. *Journal of Clinical Microbiology*, *58*(5), 1–22. https://doi.org/10.1128/JCM.00187-20

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