

Laboratory Biosafety Guidelines for Handling and Processing Specimens

1. General Guidelines (when working with potentially infectious materials)

1.1 All laboratories should perform a site-specific and activity-specific risk assessment to identify and mitigate risks. Risk assessments and mitigation measures are dependent on:

- The procedures performed
- Identification of the hazards involved in the process and/or procedures
- The competency level of the personnel who perform the procedures
- The laboratory equipment and facility
- The resources available

1.2 Follow Standard Precautions when handling clinical specimens, all of which may contain potentially infectious materials. Standard Precautions include hand hygiene and the use of personal protective equipment (PPE), such as laboratory coats or gowns, gloves, and eye protection.

1.3 Follow routine laboratory practices and procedures for decontamination of work surfaces and management of laboratory waste.

1.4 Any procedure with the potential to generate **fine-particulate aerosols** (e.g., vortex or sonication of specimens in an **open tube**) should be performed in a Class II Biological Safety Cabinet (BSC). Appropriate physical containment devices (e.g., **centrifuge safety buckets; sealed rotors**) **should be used for centrifugation.**

- a. Use parafilm to seal the tubes first and put into centrifuge cups and seal tightly before putting into the centrifuge. Wait after 15 minutes before opening the centrifuge.
- b. Ideally, rotors and buckets should be **loaded and unloaded in a BSC**. Perform any procedures outside a BSC in a manner that minimizes the risk of exposure to an inadvertent sample release.

1.5 After specimens are processed, decontaminate work surfaces and equipment and transportation box with appropriate disinfectants. Use any EPA-registered hospital disinfectant. Follow manufacturer's recommendations for use-dilution (i.e., concentration), contact time, and care in handling.

1.6 All disposable waste should be put into appropriate biohazard bag and sealed to be autoclaved or incinerated.

ANNEX 5a

2. The following activities may be performed in BSL-2 facilities using standard BSL-2 work practices: **(Normal practice)**
 - a. Pathologic examination and processing of formalin-fixed or otherwise inactivated tissues **(HPE)**
 - b. Molecular analysis of extracted nucleic acid preparations **(PCR mastermix, put into thermocycler)**
 - c. Electron microscopic studies with glutaraldehyde-fixed grids
 - d. Routine examination of bacterial and mycotic cultures **(Reading plates)**
 - e. Routine staining and microscopic analysis of fixed smears **(Staining and reading BFMP, FBP)**
 - f. Final packaging of specimens for transport to diagnostic laboratories for additional testing. Specimens should already be in a sealed, decontaminated primary container.
 - g. Inactivated specimens (e.g., specimens in nucleic acid extraction buffer/heat block)

3. The following activities involving manipulation of potentially infected specimens should be performed as above and, in a Class II BSC:
 - a. **Aliquot and/or diluting** specimens
 - b. **Inoculating** bacterial or mycological culture media **(Sample processing) PROCESS POSITIVE BLOOD CULTURES AND ANY SAMPLE FROM SUSPECTED COVID-19 CASES IN THE BSC**
 - c. Performing diagnostic tests that do not involve propagation of viral agents in vitro or in vivo
 - d. **Nucleic acid extraction** procedures involving potentially infected specimens
 - e. **Preparation and chemical- or heat-fixing of smears** for microscopic analysis. ie: AFB Smear

4. Virus Isolation

Virus isolation in cell culture and initial characterization of viral agents recovered in cultures of SARS-CoV-2 specimens should only be conducted in a Biosafety Level 3 (BSL-3) laboratory using BSL-3 practices. Site-specific and activity-specific biosafety risk assessments should be performed to determine if additional biosafety precautions are warranted based on situational needs.