Biorisk management in laboratories handling SARS-CoV-2 (COVID-19) samples

Objectives
- Providing guidelines and required biological Containment Level (CL) for handling of SARS-CoV-2 samples handling.
- Offering instructions to reducing biological risk to acceptable level and guarantee current in force law compliance.

Basic control measures

Procedures and basic primary barriers.
- Biosafety recommendations from the European Directive 54/2000/EC and WHO should be applied.
- Techniques, equipment, and Personal Protective Equipment (PPE) must be adapted to the technique performed.
- Hand-washing before entering the lab.
- Recommended PPE use in case of aerosol and splashing hazards.
  - Splash-resistant lab coat (closed at the back + elastic cuffs) or coverall, type 4B as lowest protection level.
  - Gloves for biological hazards
  - Face mask type FFP2 or higher level.
  - Anti-Splash safety goggles or face shield.
  - Disposable head covering/cap.
  - Shoe cover.
- Non-inactivated biological material must be used in certified biological safety cabinet (BSC) class II with qualification on effective recirculation.
- Aerosol – barrier microperpetes (or tips).
- Screw - capped tubes and bottles.
- Material must be disinfected before remove it from BSC.
- All BSC inner surfaces must be disinfected after work.
- Adequate programs for cleaning and disinfection and hazardous waste management.

Administrative and organizational.
- Previous, proactive and ongoing process to identify and assess Biological Risk.
- Biological Safety Officer or adviser (BSO) should be available.
- Education and training on the use of PPE, equipment and emergency and contingency procedures.
- Specific health and medical surveillance. It is advised to include diagnostic tests for COVID-19 (initial, periodical and in case of exposure suspicion) and serological proof.
- Any accident or incident must be communicated to the BSO and/or to Occupational Health personnel.

Biosecurity
- Records allowing traceability for:
  - Entry and transfers to and from entities material.
  - Inventory and location.
- Material usage.
  - Biohazard destruction and disposal.
- Safe computer records on safe location.
- Access control.
  - Regular inventory verification.
  - Immediate notification on any deviation to biosecurity officer.
- Biohazard signposting.

CL2

Laboratory techniques.
- Diagnostic techniques not implying virus culture and amplification.
  - Preparation of non-inactivated fresh samples.
  - Solid or viscous samples homogenization (sputum, bronchoalveolar lavage, …).
  - Lysis buffer addition.
  - Non-inactivated biological samples aliquoting.
  - Non-inactivated samples centrifugation.
  - Fixed samples cell sorting or Fluorescence-activated cell sorting (FACS).
  - Non-fixed samples cell sorting or FACS into BSC specifically designed or equipped with Aerosol Management System (AMS), certified and qualification on effect.

Facilities.
- Decontaminable surfaces for benchtop, floor, and walls.
- Access control, preferably electronic.
- Autoclave, available at the building.
- Recommended independent ventilation system without air recirculation.
- Continuous Inward Directional Airflow.
- On site differential pressure visual control with alarm.
- HEPA (H14) filtration on exhaust.
- Recommended HEPA (H13) filtration on inlet.
- Material exchange systems (SAS, airlock, dumb, and double door autoclave located on containment barrier).
- Changing room for clothes and PPE donning and doffing and decontamination shower at the exit.
- General efficient treatment system must be provided.
- Neutralization of biologicals generated in the laboratory before removal or discharging on general sewage system (general or local), preferably before leaving BSC.

Control measures.
- All from CL2.
- Education and training on CL3 conditions (approved by BSO).
- Personal Protective equipment:
  - Type 4B or higher coverall and boot-covers.
  - FFP3 face-mask, and/or ventilated hood.
  - Use of double glove certified “virus”.
- PPE decontamination before leaving the working area.
- Standard Operating Procedures for PPE doffing.
- Hand washing, expectation, nasal mucous cleaning and nails brushing before leaving BCL3 area.
- Validated decontamination procedures for surfaces and rooms.
- All the stocks of biological materials need to be recorded.

CL3

Laboratory techniques.
- Virus culture, isolation, purification or characterization.
- Viable cells cell-sorting or FACS with non-inactivated virus or confirmed samples.

Facilities.
- All from CL2.
- Decontaminable surfaces also for ceilings.
- Anteroom at the access to containment zone.
- No opening and sealed windows.
- Independent ventilation system without air recirculation.
  - Continuous Inward Directional Airflow.
- On site differential pressure visual control with alarm.
- HEPA (H14) filtration on exhaust.
- Recommended HEPA (H13) filtration on inlet.
- Material exchange systems (SAS, airlock, dumb, and double door autoclave located on containment barrier).
- Changing room for clothes and PPE donning and doffing and decontamination shower at the exit.
- General efficient treatment system must be provided.
- Neutralization of biologicals generated in the laboratory before removal or discharging on general sewage system (general or local), preferably before leaving BSC.

Control measures.
- All from CL2
- Education and training on CL3 conditions (approved by BSO).
- Personal Protective equipment:
  - Type 4B or higher coverall and boot-covers.
  - FFP3 face-mask, and/or ventilated hood.
  - Use of double glove certified “virus”.
- PPE decontamination before leaving the working area.
- Standard Operating Procedures for PPE doffing.
- Hand washing, expectation, nasal mucous cleaning and nails brushing before leaving BCL3 area.
- Validated decontamination procedures for surfaces and rooms.
- All the stocks of biological materials need to be recorded.

Inactivated or fixed by validated methods or with scientific evidence.

Out of CL2

Transport

External
- Diagnostic or clinical samples: Category B (UN3373).
  - Virus cultures: Category A (UN2814).
  - No private vehicles allowed.
  - Hermetic primary container.
  - BCL3: primary container must be prepared in BSC and secondary container in the lab.
  - BCL3: Both primary and secondary containers must be prepared in BSC.

Internal
- Secondary container disinfected before leaving the lab.

Control measures.
- Apply Good Microbiology Practices and Procedures (GMP).

Disinfectants

<table>
<thead>
<tr>
<th>Disinfectant</th>
<th>Concentration</th>
<th>Contact time</th>
<th>Application</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ethanol</td>
<td>70%</td>
<td>1 min</td>
<td>Surfaces</td>
</tr>
<tr>
<td>Sodium hypochlorite (bleach)</td>
<td>0.1% (1,000 ppm)</td>
<td>10 min</td>
<td>Surfaces</td>
</tr>
<tr>
<td>Sodium hypochlorite (bleach)</td>
<td>1% (10,000 ppm)</td>
<td>10 min</td>
<td>Spills or liquids with organic load</td>
</tr>
<tr>
<td>Quaternary amonium</td>
<td>Manufacturer instructions</td>
<td>Manufacturer instructions</td>
<td>Surfaces or liquids with organic load</td>
</tr>
<tr>
<td>Hydrogen peroxide – Peroxide acid</td>
<td>Manufacturer instructions</td>
<td>Manufacturer instructions</td>
<td>Surfaces or liquids with organic load</td>
</tr>
<tr>
<td>Viracide™</td>
<td>1% [1g, 1 l]</td>
<td>10 min</td>
<td></td>
</tr>
<tr>
<td>Peracide™</td>
<td>1,62% (16,2 gr, 1 l)</td>
<td>10 min</td>
<td></td>
</tr>
</tbody>
</table>

References
1. ABE BioS. Considerations for Handling Potential SARS-CoV-2 Samples. 12/05/2020
2. CCC: Interim Laboratory Biosafety Guidelines for Handling and Processing Specimens Associated with COVID-19 (sars). 31/5/2020