Laboratory biosafety guidance related to coronavirus disease (COVID-19)

Interim guidance
19 March 2020

World Health Organization

Background

The purpose of this document is to provide interim guidance on laboratory biosafety related to the testing of clinical specimens of patients that meet the case definition of the novel pathogen identified in Wuhan, China, that is, coronavirus disease 2019 COVID-19.

WHO will revise these recommendations as necessary.

Highlights of COVID-19 laboratory biosafety

- All procedures must be performed based on risk assessment and only by personnel with demonstrated capability, in strict observance of any relevant protocols at all times.
- Initial processing (before inactivation) of all specimens should take place in a validated biological safety cabinet (BSC) or primary containment device.
- Non-propagative diagnostic laboratory work (for example, sequencing, nucleic acid amplification test [NAAT]) should be conducted at a facility using procedures equivalent to Biosafety Level 2 (BSL-2).
- Propagative work (for example, virus culture, isolation or neutralization assays) should be conducted at a containment laboratory with inward directional airflow (BSL-3).
- Appropriate disinfectants with proven activity against enveloped viruses should be used (for example, hypochlorite [bleach], alcohol, hydrogen peroxide, quaternary ammonium compounds, and phenolic compounds).
- Patient specimens from suspected or confirmed cases should be transported as UN3373, "Biological Substance Category B". Viral cultures or isolates should be transported as Category A, UN2814, "infectious

Laboratory biosafety

It is essential to ensure that health laboratories adhere to appropriate biosafety practices. Any testing for the presence of the virus responsible for COVID-19 or of clinical specimens from patients meeting the suspected case definition[1] should be performed in appropriately equipped laboratories, by staff trained in the relevant technical and safety procedures. National guidelines on laboratory biosafety should be followed in all circumstances. For general information on laboratory biosafety guidelines, see the WHO Laboratory biosafety manual, 3rd edition[2] in the interim before the 4th edition is released.

Key points

- Each laboratory should conduct a local (that is, institutional) risk assessment to ensure it is competent to safely perform the intended testing with appropriate risk control measures in place.
- 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.
- The handling and processing of specimens from cases with suspected or confirmed COVID-19 infection that are intended for additional laboratory tests, such as haematology or blood gas analysis, should follow local guidelines for processing potentially infectious material.
- Non-propagative diagnostic laboratory work, including sequencing and NAAT, on clinical specimens from patients who are suspected or confirmed to be infected with COVID-19, should be conducted adopting the practices and procedures of core requirements,[a] (see Annex I), and an appropriate selection of heightened control measures,[b] as informed by the local risk assessment. In the interim, BSL-2 in the

[a] Core requirements: A set of minimum requirements defined in the 4th edition of the WHO *Laboratory biosafety manual* to describe a combination of risk control measures that are both the foundation for, and an integral part of, laboratory biosafety. These measures reflect international standards and best practice in biosafety that are necessary to work safely with biological agents, even where the associated risks are minimal.

[b] Heightened control measures: A set of risk control measures that may need to be applied in a laboratory facility because the outcome of a risk assessment indicates that the biological agents being handled and/or the activities to be performed with them are associated with a relatively high risk that cannot be acceptable solely with the core requirements.

WHO Laboratory biosafety manual, 3rd edition[2] remains appropriate until the 4th edition replaces it.

- 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 laboratories capable of meeting additional essential containment requirements and practices, that is, BSL-3.
- Initial processing (before inactivation) of all specimens, including those for sequencing and NAAT, should take place in an appropriately maintained and validated BSC or primary containment device.
- Appropriate disinfectants with proven activity against enveloped viruses should be used for the recommended contact time, at the correct dilution and within the expiry date after the working solution is prepared.
- All technical procedures should be performed in a way that minimizes the generation of aerosols and droplets.
- Appropriate personal protective equipment (PPE), as determined by a detailed risk assessment, should be worn by all laboratory personnel handling these specimens.
- Patient specimens from suspected or confirmed cases should be transported as UN3373 Biological Substance Category B. Viral cultures or isolates should be transported as Category A UN2814, infectious substance, affecting humans.[3]

Recommendations addressing minimal/essential working conditions associated with specific manipulations in laboratory settings.

The additional recommendations provided in this section address the minimal/essential working conditions associated with specific manipulations in laboratory settings.

1. Risk assessment

Risk assessment is a systematic process of gathering information and evaluating the likelihood and consequences of exposure to or release of workplace hazard(s) and determining the appropriate risk control measures to reduce the risk to an acceptable level. It is important to note that hazards alone do not pose a risk to humans or animals. Consideration therefore must also be given to the types of equipment used and the procedure(s) that will be performed with the biological agent.

It is highly recommended to start with performing a local risk assessment for each process step, that is, from sample collection, sample reception, clinical testing, polymerase chain reaction (PCR) to virus isolation (only when and where applicable). Certain hazards will then be considered for each process step, such as aerosol exposure during sample processing; eye splash during sample processing; infectious culture material spill; and leaking sample (in the case of sample reception), with an assessed grade of risk. For each identified risk, appropriate risk control measures, including but not limited to the following recommendations, should be selected and implemented, to mitigate the residual risks to an acceptable level.

A risk assessment template is provided in Annex II; this is intended to serve as an example and to facilitate the process.

2. Routine laboratory procedures, including non-propagative diagnostic work and PCR analysis.

Non-culture-based diagnostic laboratory work and PCR analysis on clinical specimens from patients who are suspected or confirmed to be infected with the virus responsible for COVID-19 should be conducted adopting practices and procedures described for conventional clinical and microbiology laboratories as described in the core requirements (see Annex I).

However, all manipulations of potentially infectious materials, including those that may cause splashes, droplets, or aerosols of infectious materials (for example, loading and unloading of sealed centrifuge cups, grinding, blending, vigorous shaking or mixing, sonic disruption, opening of containers of infectious materials whose internal pressure may be different from the ambient pressure) should be performed in appropriately maintained and validated BSCs or primary containment devices, by personnel with demonstrated capability.

Examples of routine laboratory procedures include:

- diagnostic testing of serum; blood (including haematology and clinical chemistry); respiratory specimens such as nasopharyngeal and oropharyngeal swabs, sputum and/or endotracheal aspirate or bronchoalveolar lavage; stool; or other specimens;
- routine examination of mycotic and bacterial cultures developed from respiratory tract specimens. When handling and processing specimens, core requirements (see Annex I), including GMPP, should be followed at all times, including but not limited to those under the following subheadings. More details are explained and demonstrated in the WHO Biosafety video series.[4]

3. Use of appropriate disinfectants

While little is known about this novel virus, the comparable genetic characteristics between the virus responsible for COVID-19 and MERS-CoV suggest that the COVID-19 virus may be susceptible to disinfectants with proven activity against enveloped viruses, including sodium hypochlorite (bleach; for example, 1000 parts per million [ppm] (0.1%) for general surface disinfection and 10 000 ppm (1%) for disinfection of blood spills); 62-71% ethanol; 0.5% hydrogen peroxide; quaternary ammonium compounds; and phenolic compounds, if used according to the manufacturer's recommendations. Other biocidal agents such as 0.05–0.2% benzalkonium chloride or 0.02% chlorhexidine digluconate can be less effective.

Particular attention should be paid not only to the selection of the disinfectant but also the contact time (for example, 10 minutes), dilution (that is, concentration of the active ingredient) and expiry date after the working solution is prepared.

Human coronaviruses in general are known to persist on inanimate surfaces such as metal, glass or plastic for up to a maximum of 9 days.[5]

4. Viral isolation

Unless a country decides otherwise, considering the newly acquired knowledge and effective preventive measures described above, viral isolation on clinical specimens from patients who are suspected or confirmed to be infected with the virus responsible for COVID-19 should be performed only in laboratories capable of meeting the following additional containment criteria:

- a controlled ventilation system maintains inward directional airflow into the laboratory room;
- exhaust air from the laboratory room is not recirculated to other areas within the building. Air must be HEPA (high-efficiency particulate air) filtered, if reconditioned and recirculated within the laboratory. When exhaust air from the laboratory is discharged to the outdoors, it must be dispersed away from occupied buildings and air intakes. This air should be discharged through HEPA filters;
- a dedicated hand-wash sink is available in the laboratory;
- all manipulations of infectious or potentially infectious materials must be performed in appropriately maintained and validated BSCs;
- laboratory workers should wear protective equipment, including disposable gloves; solid-front or wrap-around gowns, scrub suits, or coveralls with sleeves that fully cover the forearms; head coverings; shoe covers or dedicated shoes; and eye protection (goggles or face shield). Risk assessment should inform the use of respiratory protection (fit-tested particulate respirator, for example, EU FFP2, US 6 NIOSH-certified N95 or equivalent, or higher protection);
- centrifugation of specimens should be performed using sealed centrifuge rotors or sample cups. These rotors or cups should be loaded and unloaded in a BSC.

5. Additional risks associated with virus isolation studies.

Certain experimental procedures may carry additional risks of virus mutations with possible increased pathogenicity and/or transmissibility, or viruses with altered antigenicity or drug susceptibility. Specific risk assessments should be conducted, and specific risk-reduction measures adopted, before any of the following procedures are conducted:

- coinfection of cell cultures with different coronaviruses, or any procedures that may result in a coinfection;
- culture of viruses in the presence of antiviral drugs;
- deliberate genetic modification of viruses.

6. Work with animals infected with the virus responsible for COVID-19.

The following activities require an animal facility – BSL-3 facilities and work practices, as detailed in the WHO Laboratory biosafety manual, 3rd edition:[2]

- inoculation of animals for potential recovery of the agent from specimens of the virus responsible for COVID-19;
- any protocol involving animal inoculation for confirmation and/or characterization of putative agents of the COVID-19 virus.

7. Referral of specimens to laboratories with appropriate containment measures in place.

Laboratories that are not able to meet the above biosafety recommendations should consider transferring specimens to national, regional, or international referral laboratories with COVID-19-detection capacity that can meet the biosafety requirements.

Packaging and shipment

All materials transported within and between laboratories should be placed in a secondary container, to minimize the potential for breakage or a spill. An example includes transfer of materials from the BSC to an incubator and vice versa. Specimens leaving the BSC should be surface decontaminated. Detailed guidance is provided in the WHO Biosafety video series,[4] in particular, Good microbiological practices and procedures (GMPP) 7: transport.

Transport of specimens within national borders should comply with national regulations. Cross-boundary transport of specimens of the virus responsible for COVID-19 should follow the United Nations model regulations, Technical instructions for the safe transport of dangerous goods by air (Doc 9284) of the International Civil Aviation Organization,[6] for airlifted transport, and any other applicable regulations depending on the mode of transport being used.

More information may be found in the WHO Guidance on regulations for the transport of infectious substances 2019-2020 (applicable as from 1 January 2019).[3] A summary on transport of infectious substances can also be found in Tool box 4 of the WHO handbook, Managing epidemics: key facts about deadly diseases.[7]

Patient specimens from suspected or confirmed cases should be transported as UN3373, Biological Substance Category B, when they are transported for diagnostic or investigational purposes. Viral cultures or isolates should be transported as Category A UN2814, infectious substance, affecting humans.[3]All specimens being transported (whether UN3373 or UN2814) should have appropriate packaging, labelling, and documentation, as described in the documents mentioned earlier.

References

1. World Health Organization. Global Surveillance for human infection with coronavirus disease (COVID-19) (accessed 17 February 2020).
2. Laboratory biosafety manual, 3rd ed. Geneva: World Health Organization; 2004 (https://www.who.int/csr/resources/publications/biosafety/Biosafety7.pdf?ua=1, accessed 14 February 2020).
3. Guidance on regulations for the transport of infectious substances 2019–2020. Geneva: World Health Organization; 2019 (WHO/WHE/CPI/2019.20; https://www.who.int/ihr/publications/WHO-WHE-CPI-2019.20/en/, accessed14 February 2020).
4. World Health Organization. Strengthening health security by implementing the International Health Regulations (2005). Biosafety video series (https://www.who.int/ihr/publications/biosafety-video-series/en/, accessed 14 February 2020).
5. Kampf G, Todt D, Pfaender S, Steinmann E. Persistence of coronaviruses on inanimate surfaces and its inactivation with biocidal agents. J Hosp Infect. 2020;Feb 6. Pii:s0195-6701(20)30046-3. doi:10.1016/j.jhin.2020.01.022 [epub ahead of print].
6. International Civil Aviation Organization (ICAO). Safety. Technical instructions for the safe transport of dangerous goods by air (Doc 9284) (https://www.icao.int/safety/DangerousGoods/Pages/technical-instructions.aspx, accessed 14 February 2020).
7. Managing epidemics: key facts about deadly diseases. Geneva: World Health Organization; 2018 (https://apps.who.int/iris/handle/10665/272442, accessed 14 February 2020).
8. How to handrub? With alcohol-based formulation. How to handwash? With soap and water. Geneva: World Health organization; 2006 (https://www.who.int/gpsc/tools/GPSC-HandRub-Wash.pdf, accessed 15 February 2020).

Acknowledgements

The following people contributed to the current revision of this guidance: Stuart Blacksell, Mahidol Oxford Tropical Medicine Research Unit, Thailand; Kathrin Summermatter, Institute for Infectious Diseases, University of Bern, Switzerland.

WHO Health Emergency Programme: Kazunobu Kojima, Rica Zinsky, Zsofia Igloi.

WHO continues to monitor the situation closely for any changes that may affect this interim guidance. Should any factors change, WHO will issue a further update. Otherwise, this interim guidance document will expire 2 years after the date of publication.

Annex I: Core requirements

1. Good microbiological practice and procedure (GMPP).

Best practice

• Never store food or drink, or personal items such as coats and bags in the laboratory. Activities such as eating, drinking, smoking, and applying cosmetics are only to be performed outside the laboratory.

• Never put materials, such as pens, pencils or gum in the mouth while inside the laboratory, regardless of having gloved hands or not.

• Thoroughly wash hands *(8)*, preferably with warm running water and soap, after handling any biological material, including animals, before leaving the laboratory, and any time contamination is known or suspected to be present on the hands.

• Ensure open flames or heat sources are never placed near flammable supplies and are never left unattended.

• Ensure that coverings are placed over any cuts or broken skin prior to entering the laboratory.

• Ensure, prior to entry into the laboratory, that supplies of laboratory equipment and consumables, including reagents, PPE and disinfectants, are sufficient and appropriate for the activities being performed.

• Ensure supplies are stored appropriately (that is, according to storage instructions) and safely, to reduce the chance of accidents and incidents such as spills, trips or falls for laboratory personnel.

• Ensure proper labelling of all biological agents and chemical and radioactive material.

• Protect written documents from contamination using barriers (such as plastic coverings), particularly those that may need to be removed from the laboratory.

• Ensure work is performed with care, in a timely manner and without rushing. Working when fatigued should be avoided.

• Keep the work area tidy, clean, and free of clutter and materials that are not necessary for the work being done.

• Prohibit the use of earphones, which can distract personnel and prevent equipment or facility alarms from being heard.

• Appropriately cover or remove any jewellery that could tear glove material, easily become contaminated or act as a fomite for infection. If worn regularly, cleaning and decontamination of jewellery or spectacles should be considered.

• Refrain from using mobile electronic devices (for example, mobile telephones, tablets, laptops, flash drives, memory sticks, cameras, or other portable devices, including those used for DNA/RNA sequencing) when not specifically required for the laboratory procedures being performed.

• Keep mobile electronic devices in areas where they could not easily become contaminated or act as a fomite for infection. Where close proximity of such devices to biological agents is unavoidable, ensure they are either protected by a physical barrier or decontaminated before leaving the laboratory.

Technical procedures

• Avoid inhalation of biological agents. Use good techniques to minimize the formation of aerosols and droplets when manipulating specimens.

• Avoid ingestion of biological agents and contact with the skin and eyes.

• Wear disposable gloves at all times when handling specimens.

• Avoid contact of gloved hands with the face.

• Shield or otherwise protect the mouth, eyes, and face during procedures where splashes may occur.

• Wherever possible, replace any glassware with plasticware.

• For work requiring scissors, use scissors with blunt or rounded ends in preference to those with pointed ends.

• Handle all sharps, syringes, and needles, if necessary, with care so as to prevent injury and injection of biological agents.

• Use ampoule openers for safe handling of ampoules.

• Never re-cap, clip, or remove needles from disposable syringes.

• Dispose of any sharps materials (for example, needles, needles combined with syringes, blades, broken glass) in puncture-proof or puncture-resistant containers fitted with sealed covers.

• Preventing dispersal of biological agents:
  – discard specimens and cultures for disposal in leak-proof containers with the tops appropriately secured before disposal in dedicated waste containers;
  – consider opening tubes with disinfectant-soaked pad/gauze;
  – decontaminate work surfaces with a suitable disinfectant at the end of the work procedures and if any material is spilled or obviously contaminated;
  – ensure the disinfectant is efficacious against the pathogen being handled and is left in contact with infectious waste materials for sufficient time to effect complete inactivation.

2. Personnel competence and training

General familiarization and awareness training

General training should include an introduction to laboratory layout, codes of practice, local guidelines, safety manuals, risk assessments, legislative requirements, and emergency response procedures.

Job-specific training
• Training requirements may vary depending on the job functions.

• However, in general, all personnel involved in the handling of biological agents must be trained on GMPP.

• Competency and proficiency assessment must be used and verified before working independently, followed by regular review and refresher training.

• Relevant information such as new procedures must be updated and communicated to applicable personnel.

Safety and security training
• All personnel must be aware of hazards present in the laboratory and their associated risks; safe working procedures; security measures; and emergency preparedness and response.

3. Facility design

• Ample space and a designated hand-washing basin must be provided, with appropriate restriction of access.

• Doors must be appropriately labelled, and laboratory walls, floors, and furniture must be smooth, easy to clean, impermeable to liquids and resistant to the chemicals and disinfectants normally used in the laboratory.

• Laboratory ventilation, where provided (including heating/cooling systems and especially fans/local cooling split-system air-conditioning units – specifically when retrofitted) should ensure airflows do not compromise safe working. Consideration must be made for resultant airflow speeds and directions, and turbulent airflows should be avoided; this applies also to natural ventilation.

• Laboratory space and facilities must be adequate and appropriate for safe handling and storage of infectious and other hazardous materials, such as chemicals and solvents.

• Facilities for eating and drinking must be provided outside the laboratory, and first-aid-facilities must be accessible.

• Appropriate methods for decontamination of waste, for example disinfectants and autoclaves, must be available close to the laboratory.

• The management of waste must be considered in the laboratory design. Safety systems must cover fire, electrical emergencies, and emergency/incident response facilities, based on risk assessment.

• There must be a reliable and adequate electricity supply and lighting to permit safe exit.

• Emergency situations must be considered in the design, as indicated in the local risk assessment, and should include the geographical/meteorological context.

4. Specimen receipt and storage

• A specimen received by the laboratory must be accompanied by sufficient information to identify what it is, when and where it was taken or prepared, and which tests and/or procedures (if any) are to be performed.

• Consider unpacking the items in the BSC. Personnel unpacking and receiving specimens must be adequately trained in awareness of the hazards involved; how to adopt necessary precautions according to GMPP described earlier; how to handle broken or leaking containers; and how to handle spills and use disinfectants to manage any contamination.

• Specimens must be stored in containers with adequate strength, integrity, and volume to contain the specimen; leakproof when the cap or stopper is correctly applied; made of plastic whenever possible; free of any biological material on the outside of the packaging; correctly labelled, marked, and recorded to facilitate identification; and made of an appropriate material for the type of storage required.

• Inactivation methods must be appropriately validated whenever an inactivation step is used, before transferring the specimens to other areas for further manipulation, such as PCR analysis.

5. Decontamination and waste management

• Any surface or material known to be, or potentially be, contaminated by biological agents during laboratory operations must be correctly disinfected to control infectious risks.

• Proper processes for the identification and segregation of contaminated materials must be adopted before decontamination or disposal.

• Where decontamination cannot be performed in the laboratory area or onsite, the contaminated waste must be packaged in an approved (that is, leakproof) manner, for transfer to another facility with decontamination capacity.

6. Personal protective equipment

• Laboratory coats must be used in laboratories to prevent personal clothing from getting splashed or contaminated by biological agents. Laboratory coats must have long sleeves, preferably with elasticated or fitted cuffs, and must be worn closed. Sleeves should never be rolled up. Coats must be long enough to cover the knees, but not trail on the floor. They should be fastened when worn in the laboratory. Where possible, the fabric of the laboratory coat should be splash-resistant and overlap to provide a solid front. Laboratory coats must only be worn in designated areas. When not in use, they should be stored appropriately; they should not be hung on top of other laboratory coats, or in lockers or on hooks with personal items.

• Appropriate disposable gloves must be worn for all procedures that may involve planned or inadvertent contact with blood, body fluids or other potentially infectious materials. They must not be disinfected or reused, as exposure to disinfectants and prolonged wear will reduce the integrity of the glove and decrease protection to the user. Gloves should always be inspected before use, to check they are intact.

• Safety glasses, safety goggles, face shields (visors) or other protective devices must be worn whenever it is necessary to protect the eyes and face from splashes, impacting objects, or artificial ultraviolet radiation. Eye protection can be reused but must be regularly cleaned after every use. If splashed, it must be decontaminated with an appropriate disinfectant.

• Footwear must be worn in the laboratory and must be of a design that minimizes slips and trips and can reduce the likelihood of injury from falling objects and exposure to biological agents.

• Respiratory protection is generally not a part of the core requirements. In this particular context, however, a local risk assessment should be conducted to determine whether the use of respiratory protection is needed, especially when procedures that may create aerosols and droplets will be performed outside the BSC, for example, centrifugation, handling leaking samples and procedures that can cause splashes (for example, loading and unloading of sealed centrifuge cups, grinding, blending, vigorous shaking or mixing, sonic disruption, opening of containers of infectious materials whose internal pressure may be different from the ambient pressure).

7. Laboratory equipment

• When used effectively together with GMPP, the safe use of laboratory equipment will help to minimize the likelihood of exposure of personnel when handling or manipulating biological agents.

• For equipment to effectively reduce risks, laboratory management must make sure sufficient space is provided for its use. An appropriate budget must be available for the equipment's operation and maintenance, including equipment incorporated into the facility design, which should be accompanied by specifications that outline its safety features. All personnel operating or maintaining a piece of equipment must be properly trained and be able to demonstrate proficiency.

8. Emergency/incident response plan

• Even when carrying out low-risk work and following all core requirements for biosafety, incidents can still occur. To reduce the likelihood of exposure to/release of a biological agent, or to reduce the consequences of such incidents, a contingency plan must be developed that provides specific standard operating procedures (SOPs) to be followed in possible emergency scenarios that apply to the work and local environment. Personnel must be trained on these procedures and have periodic refresher training to maintain competency.

• First-aid kits, including medical supplies such as bottled eye washes and bandages, must be available and easily accessible to personnel. These must be checked routinely to make sure products are within their use-by dates and are in sufficient supply.

• All incidents must be reported to the appropriate personnel in a timely manner. A written record of accidents and incidents must be maintained, in line with national regulations where applicable. Any incident must be reported and investigated in a timely manner and used for updating laboratory procedures and emergency response plans.

• Spill kits, including disinfectant, must be easily accessible to personnel. Depending on the size, location, concentration, or volume of the spill, different protocols may be necessary. Written procedures for cleaning and decontaminating spills must be developed for the laboratory and followed by suitably trained personnel.

9. Occupational health

• The employing authority, through the laboratory director, must take responsibility for ensuring that the health of laboratory personnel is adequately checked and reported.

• Medical examination or health status information of laboratory personnel may be required to ensure that it is safe for them to work in the laboratory.