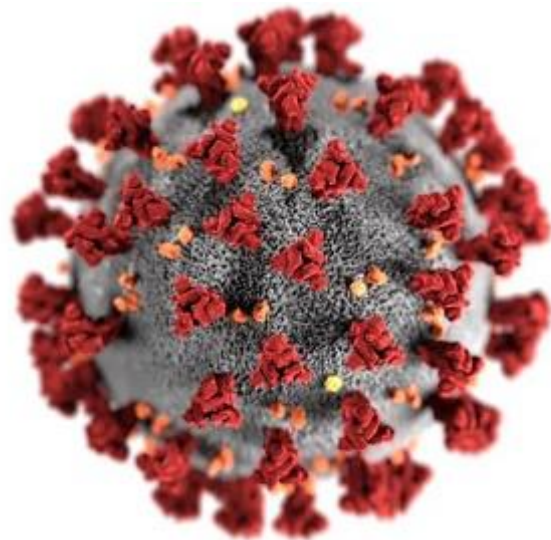


COVID-19 laboratory biosafety: elaborations on the latest WHO Laboratory biosafety guidance related to COVID-19

EURO Webinar, 12 November 2020



Kazunobu Kojima, WHO HQ



World Health Organization



Overview of Key Points



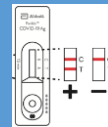
Good Microbiological Practices and Procedures



Biological Risk Assessment



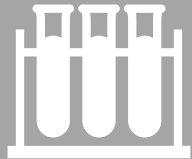
Clinical testing (non-propagative)



Antigen rapid diagnostic test (Ag-RDT)



Disinfection, inactivation, waste management



Good Microbiological Practices and Procedures

Good Microbiological Practice and Procedure (GMPP)

A series of best biosafe practices and procedures for working with infectious material in the laboratory

- Hand hygiene
- Prevent dispersal
 - Appropriate decontamination and deactivation/disposal
- Avoid injection
 - Safe sharp procedures
- Avoid ingestion and contact with skin and eyes
 - Use PPE
- Avoid inhalation
 - Prevent aerosol formation



GMPP is part of the “Core Requirements” (see Annex I of the Laboratory biosafety guidance related to coronavirus disease (COVID-19))

<https://www.who.int/ihr/publications/biosafety-video-series/en/>

[https://www.who.int/publications/i/item/laboratory-biosafety-guidance-related-to-coronavirus-disease-\(covid-19\)](https://www.who.int/publications/i/item/laboratory-biosafety-guidance-related-to-coronavirus-disease-(covid-19))



Biological Risk Assessment

Biological Risk Assessment

A systematic process of gathering information and evaluating the likelihood and impact of exposure to or release of workplace hazard(s) and determining the appropriate risk control measures to reduce the risk.

STEP 1. Gather information (hazard identification)

STEP 2. Evaluate the risks

STEP 3. Develop a risk control strategy

STEP 4. Select and implement risk control measures

STEP 5. Review risks and risk control measures



Refer to Annex II of the WHO Interim Guidance

This process is best carried out by a team of staff that are involved in various processes related to the laboratory work



STEP 1. Gather information (hazard identification)

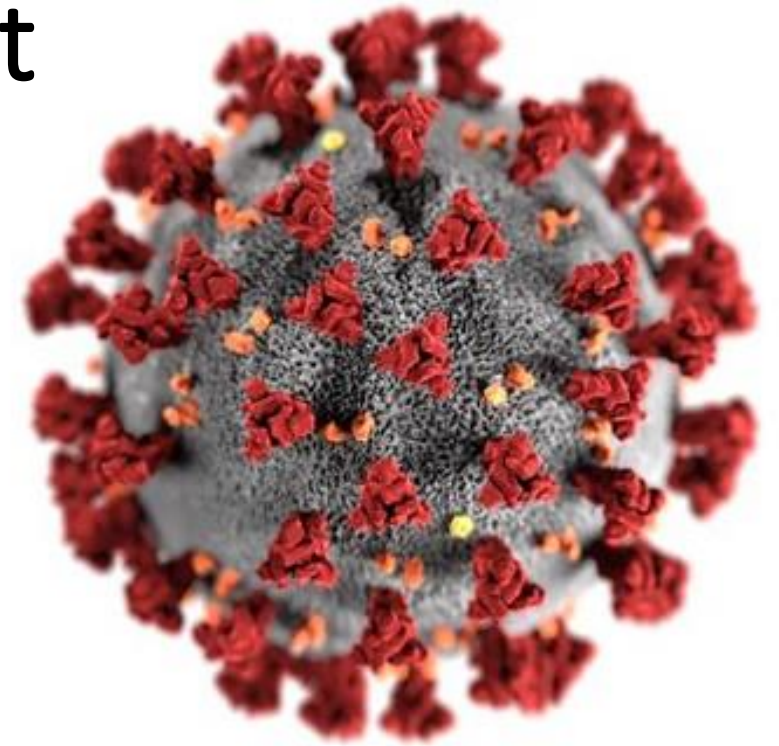
Consider the laboratory process to be performed and the following factors that influence risk:

- The biological agent (SARS-CoV-2)
- Laboratory procedures and equipment
- Control measures already in place
- Facility
- Personnel
- Other factors



SARS-CoV-2: the aetiological agent

- Transmitted via aerosolized droplets (possibly aerosols)
- No vaccine
- Highly contagious
- Infectious dose unknown
- Surface half-life uncertain
- Non-specific and varied symptoms
- Asymptomatic persons can spread disease
- Severe morbidity among immuno-incompetent and some persons with comorbidities
- Likelihood of mortality increases with age and infirmity
- No preexisting specific immunity in human population
- Some antiviral drugs under trial; treatment of symptoms

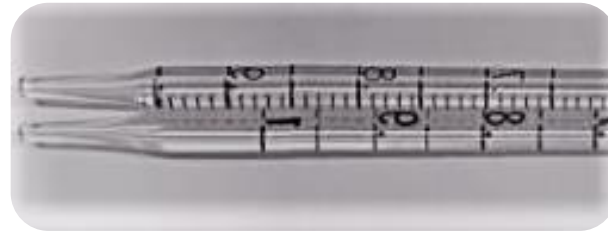


These factors will influence the consequence of accidental exposure or release!

Procedures and equipment

Aerosol producing procedures:

- Vortexing
- Shaking
- Centrifuging
- Pipetting



Sharps use (glass or needles)

Culture – highly concentrated or large volumes of virus

Laboratory animals - scratches or bites



These procedures increase the likelihood of an accidental exposure or release

Control measures in place

Biocontainment

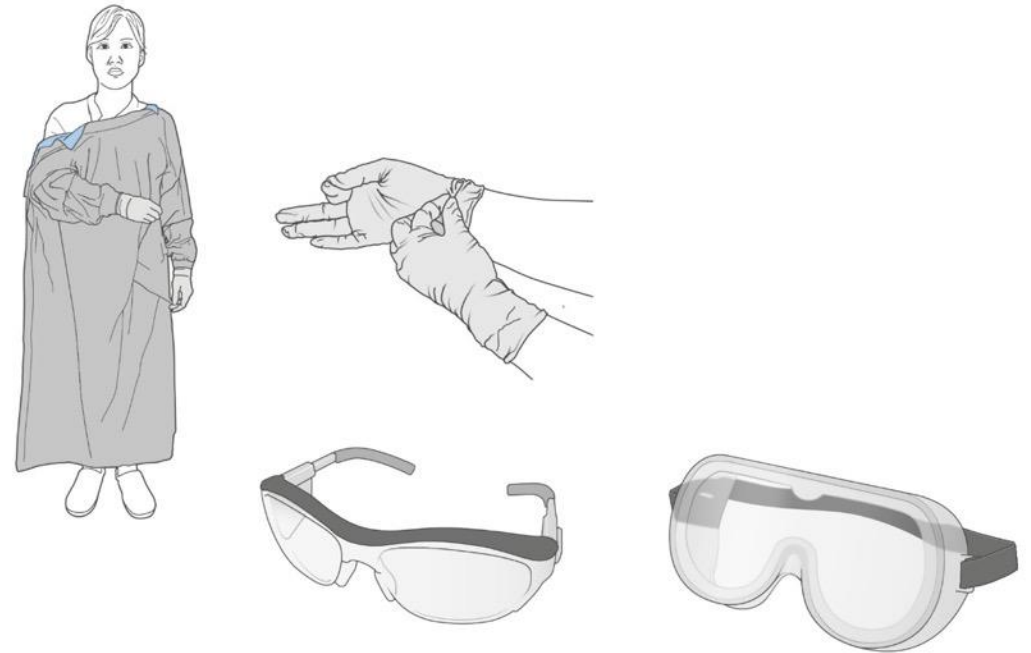
- Biosafety cabinet (BSC)
- Glovebox (possible alternative)

Personal Protective Equipment

- Disposable gloves
- Full-length laboratory coats/gowns
- Eye protection
- Face shields
- Masks/respirators

Administrative Controls

- Training
- Good Microbiological Practice and Procedure (GMPP)
- Standard operating procedures (SOPs)
- Biosafety manual



These control measures reduce the likelihood of an accidental release or exposure

Facility

Integrity

- Ample space with a hand-washing basin
- Intact (no gaps or breaches in structure)
 - Easy to clean and decontaminate
- Designed or refitted for safe, efficient and ergonomic operations

Safety and Security

- Restricted access to labs/corridors
- Doors labelled with biohazard sign
- Workflow – tidy and uncluttered

Ventilation

- Sufficient ventilation
- Directional airflow into the lab (virus isolation)



Facilities with these features reduce the likelihood of an accidental release or exposure

Personnel

Competence

Trained to perform the work

- Methods and equipment
- Biosafe practices and correct use of PPE
- Continual learning

Understanding of risks

- Mitigation and remediation

Experience

- Trained and knowledgeable in relevant lab techniques

Attitude

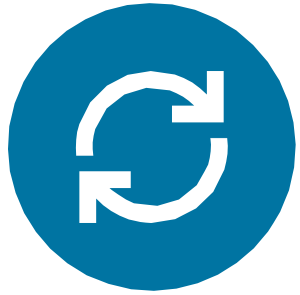
- Professional
- Focused



Well-trained, experienced laboratory personnel reduce the likelihood of an accidental release or exposure

Adding control measures to reduce risk

Procedures	Hazards	Inherent Risk	Additional Control Measures	Residual risk
Sample collection*	<ul style="list-style-type: none"> Aerosol exposure during sample processing Eye splash during sample processing Infectious material spill 	Medium to High	Face shield, respiratory protection	Low to Medium
Viral Culture*		Medium to High	Heightened containment/BSL3, inward air flow, BSC, enhanced respiratory protection	Low to Medium
RT-PCR ELISA (serology)		Medium	BSC, respiratory protection, eye protection, ventilation	Low
Near POC		Low to Medium	Respiratory protection, eye protection or face shield, ventilation	Low
POC		Low	Respiratory protection, eye protection or face shield, ventilation	Low
Sample accessioning	<ul style="list-style-type: none"> Container leaks Container breakage (sharps) Infectious material spill 	Low to Medium	Respiratory protection, eye protection or face shield, ventilation	Low
Whole Genome Sequencing	None	Very low	None needed	Very low



STEP 5. Review risks and risk control measures

- Risk assessment should be a continuous process
- Should be performed whenever changes take place:
 - Personnel
 - Facility
 - Equipment
 - Methods
 - Regulations





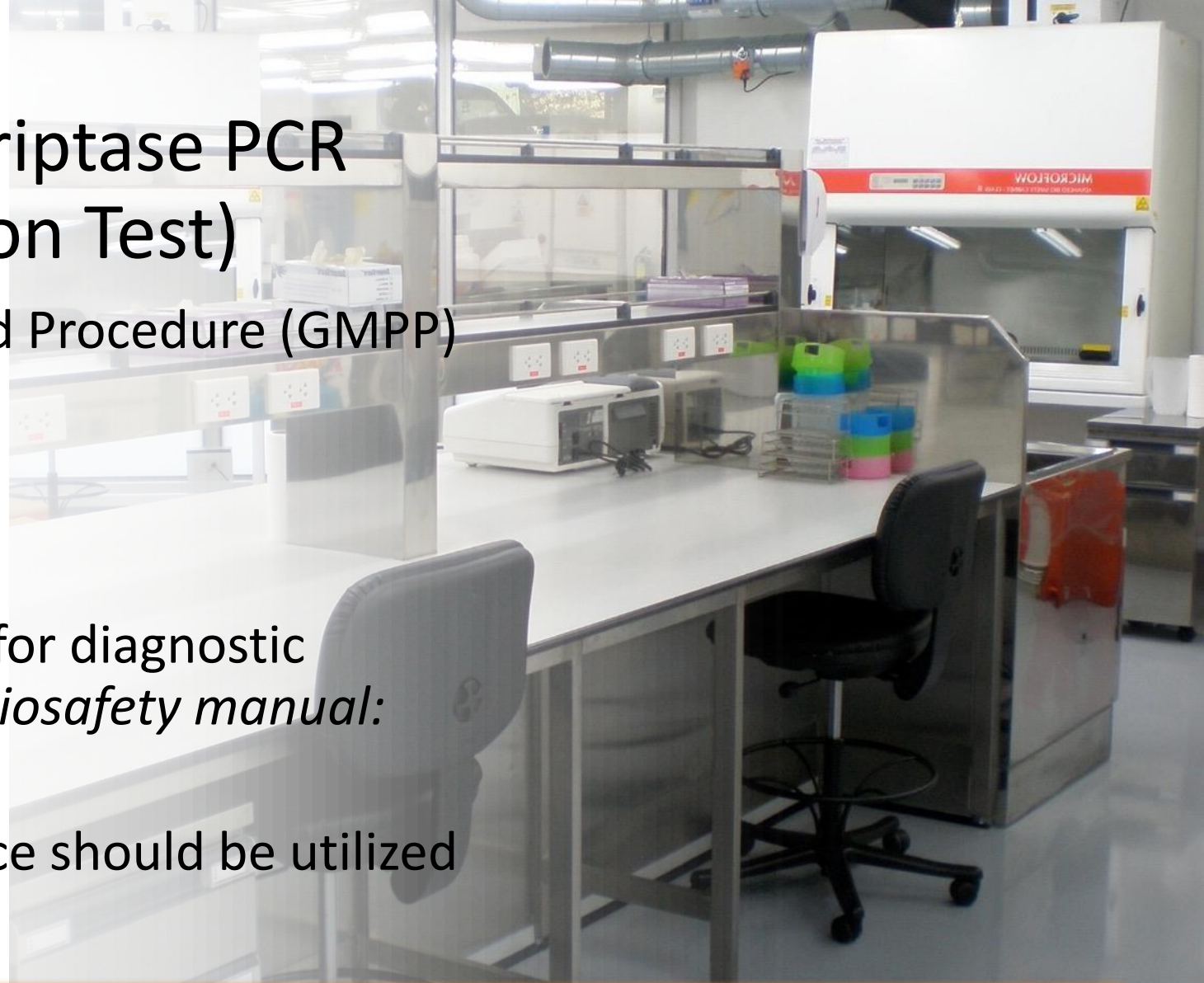
Clinical testing (non-propagative)

Real Time Reverse Transcriptase PCR (Nucleic Acid Amplification Test)

- Good Microbiological Practice and Procedure (GMPP)
 - (See “Core Requirements”, Annex I)
- Appropriate PPE
- Staff Competence
- Biosafety Level 2 (BSL-2) suitable for diagnostic services in the WHO *Laboratory biosafety manual: third edition*
- BSC or primary containment device should be utilized

External lysis buffer of the common RNA extraction kits is effective in inactivating the COVID-19 virus without heat or other additional means

<https://www.fda.gov/media/134922/download>



Point of Care (PoC) and near-POC Assays

including antigen-detecting RDTs (Ag-RDT)
(No nucleic acid extraction)

- Good Microbiological Practice and Procedure (GMPP)
- Appropriate PPE
- Staff Competence
- May be performed on bench (outside a lab)
 - Well-ventilated area (see the following slides)
 - On absorbent towel or diaper
 - Free of clutter
- Optional
 - Biosafety cabinet/glove box
 - Use primary containment if readily available

<https://www.fda.gov/media/134922/download>



Ventilation

The movement of fresh air around a closed space, or the system that does this

Types

- Natural:

Purpose-built, building openings (windows, doors, whirlybirds, chimneys, etc.)

- Assisted (mixed mode):

Relies on natural driving forces to provide the desired (design) flow rate.

- Mechanical- Fans drive mechanical ventilation.

Installed in windows, walls, air ducts



Management decides the type of lab ventilation based on suitability and availability



Disinfection, inactivation, waste management

Disinfection

1. Sodium hypochlorite (bleach)¹

- 1000 parts per million [ppm] (0.1%) for general surface disinfection
- 10 000 ppm (1%) for disinfection of sample spills
- Prepare new dilution every 24 hours
- Contact time \geq 10 min

2. Ethanol (EtOH) 62–71% (Contact time \geq 10 min)

3. Hydrogen peroxide (H₂O₂) 0.5%

4. Quaternary ammonium compounds and phenolic compounds, if used according to the manufacturer's recommendations

5. Other compounds according to manufacturer's directions²

- Use with caution in well-ventilated areas
- Allow appropriate contact time
- Do not use expired chemicals



1. <https://www.who.int/csr/resources/publications/biosafety/Biosafety7.pdf?ua=1>

2. <https://www.epa.gov/pesticide-registration/list-n-disinfectants-use-against-sars-cov-2-covid-19>

Inactivation

1. Chemical

- Some viral RNA extractions buffers^{1,2,3}
- Formalin for tissue samples³

2. Gamma Irradiation (≥ 1 Mrad)⁴

3. Heat

- 30 min at 65°C² (conservative)
- *Serology – may be affected
(Read manufacturer's instructions)



Inactivate SARS CoV-2 whenever possible BEFORE manipulation to prevent accidental exposure or release

1. <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7354533/pdf/viruses-12-00624.pdf>

2. <https://www.fda.gov/media/134922/download>

3. <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7354523/pdf/viruses-12-00622.pdf>

4. <https://absa.org/wp-content/uploads/2020/04/ABSA2020-InVitroInactivation-ofSARS-CoV-2-UsingGammaRadiation.pdf>

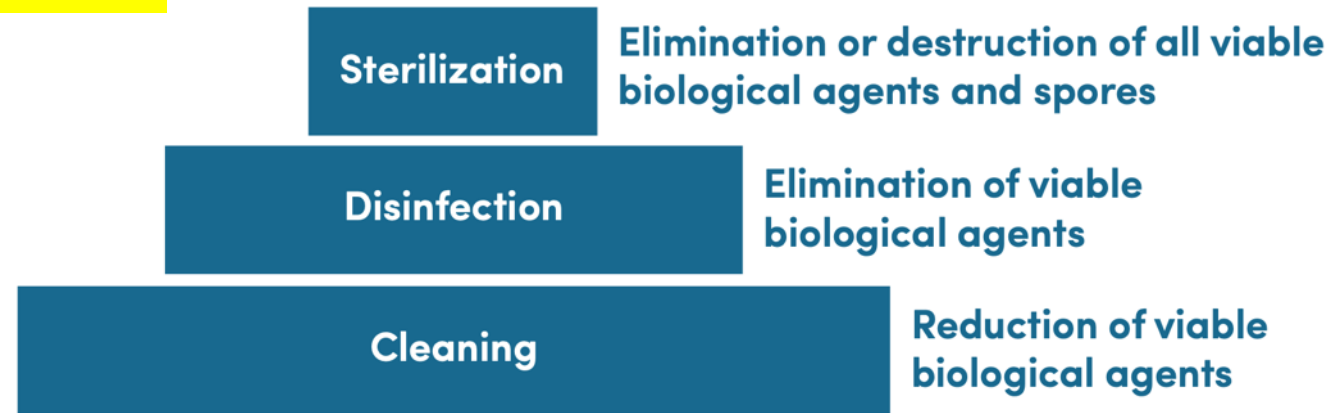
Decontamination and waste management principles

1. Control biological risks

- Any surfaces or materials known to be or potentially contaminated
- Benchtops, interior surfaces of BSC, equipment and devices

2. Identify and Segregate contaminated materials

- Sharps
- Contaminated waste
- Chemical waste
 - e.g. Sodium azide (Ag-RDT buffer)
 - Toxic and explosive gas in plumbing systems
- General (non-hazardous) waste



3. For all contaminated materials or liquids

- Decontaminate onsite to allow further safe handling *or* package and transport safely to another treatment site

Waste Management

- Autoclave or incinerate infectious waste¹
- Waste is Category B for transportation purposes
 - Regulated Medical Waste UN 3291
- Disposal of POC spent test cartridges
 - Read manufacturers specific instructions
 - Read Material Safety Data Sheets
 - Follow national, local regulations for disposal



1. <https://www.who.int/csr/resources/publications/biosafety/Biosafety7.pdf?ua=1>

Remember...

Use caution when working with products containing guanidinium iso/thiocyanate (GITC/GTC)

GTC lyses cells and denatures nucleases (RNase/DNase)

Products containing GITC

- Most DNA/RNA extraction kits
- GeneXpert cartridges
- TRIzol™ and similar products
- *Some* viral transport media (e.g. PrimeStore® MTM, Zymo DNA/RNA Shield)

Read and follow manufacturer's instructions and Safety Data Sheets (SDS/MSDS)

Do not use bleach in the presence of GTC

- **Reaction produces cyanide and chlorine gases**
- GTC inactivates organisms, so bleach not required

GTC waste is Hazardous Waste

- Toxic to marine and aquatic life
- **Do not** dispose of in wastewater stream
- Segregate GTC waste
 - Dispose of according to federal, state and local guidelines

Reference, Acknowledgements, Thanks

WHO Laboratory biosafety guidance related to coronavirus disease (COVID-19) Interim guidance 13 May 2020

[https://www.who.int/publications/i/item/laboratory-biosafety-guidance-related-to-coronavirus-disease-\(covid-19\)](https://www.who.int/publications/i/item/laboratory-biosafety-guidance-related-to-coronavirus-disease-(covid-19))

- Christina Scheel (Centers for Disease Control and Prevention, United States of America)
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