

Inactivation methods for SARS-CoV-2

Viral inactivation is an important step prior to reducing the biocontainment level for handling materials that contain SARS-CoV-2. Viral inactivation destroys the ability of the virus to replicate but seeks to preserve viral and host proteins or nucleic acid.

Inactivation methods include chemical treatment (detergents, fixatives, denaturants), physical treatment (heat, ionizing radiation, UV light) or a combination of the two. Kit reagents can come with a validated claim that they inactivate enveloped viruses. Freezing will not inactivate virus.

Influence of specimen matrix and reagent combinations

When an investigator seeks EH&S approval for an inactivation method, biosafety officers and subject matter experts on committees review each method and any supporting references or claims that the investigator provides. Each method is reviewed on its own merits for the matrix that the virus is present in. For example chemical fixation times may be different for tissues versus isolated cells. Cells in serum may require more rigorous inactivation conditions than cells in buffered saline. Similarly, ionic and non-ionic detergents plus heat may be more effective at inactivating virus in combination than alone.

Inactivation protocol submission

Investigators submitting protocols as a Rascal Appendix A should specifically describe inactivation reagents, concentrations, length of treatment, temperature and specimen types. EH&S maintains a list of methods that have been reviewed and deemed effective. These include chemical fixatives (histology), guanidinium isothiocyanate (RNA extraction), detergents (protein assays), and methanol (mass spec.)

Kit reagents - acceptable

CDC has published a list of kit lysis buffers for RNA isolation that inactivate SARS-CoV-2 when used according to the manufacturer's instructions (table reproduced on next page).

EH&S has also approved the following nucleic acid isolation kits:

DNA/RNA shield (Zymo Research)
Oragene CP-190 Saliva collection kit (DNAgenotek)

Kit reagents - unacceptable

These nucleic acid preservation kits do not have demonstrated claims to inactivate SARS-CoV-2.

RNA later (Qiagen)
Omnigene Oral OM-501 (DNAgenotek)

RNA Extraction Options

For each of the kits listed below, CDC has confirmed that the external lysis buffer is effective for inactivation of SARS-CoV-2.

Instrument/Manufacturer	Extraction Kit	Catalog No.
QIAGEN	² QIAamp DSP Viral RNA Mini Kit	50 extractions (61904)
	² QIAamp Viral RNA Mini Kit	50 extractions (52904) 250 extractions (52906)
QIAGEN EZ1 Advanced XL	² EZ1 DSP Virus Kit	48 extractions (62724) Buffer AVL (19073) EZ1 Advanced XL DSP Virus Card (9018703)
	² EZ1 Virus Mini Kit v2.0	48 extractions (955134) Buffer AVL (19073) EZ1 Advanced XL Virus Card v2.0 (9018708)
¹ Roche MagNA Pure LC	² Total Nucleic Acid Kit	192 extractions (03 038 505 001)
¹ Roche MagNA Pure Compact	² Nucleic Acid Isolation Kit I	32 extractions (03 730 964 001)
¹ Roche MagNA Pure 96	² DNA and Viral NA Small Volume Kit	576 extractions (06 543 588 001) External Lysis Buffer (06 374 913 001)
¹ QIAGEN QIAcube	² QIAamp DSP Viral RNA Mini Kit	50 extractions (61904)
	² QIAamp Viral RNA Mini Kit	50 extractions (52904) 250 extractions (52906)
^{1, 3} bioMérieux NucliSENS [®] easyMAG [®] and ^{1, 3} bioMérieux EMAG [®] (Automated magnetic extraction reagents sold separately. Both instruments use the same reagents and disposables, with the exception of tips.)		EasyMAG [®] Magnetic Silica (280133) EasyMAG [®] Lysis Buffer (280134) EasyMAG [®] Lysis Buffer, 2 mL (200292) EasyMAG [®] Wash Buffers 1,2, and 3 (280130, 280131, 280132) EasyMAG [®] Disposables (280135) Biohit Pipette Tips (easyMAG [®] only) (280146) EMAG [®] 1000µL Tips (418922)

¹Equivalence and performance of these extraction platforms for extraction of viral RNA were demonstrated with the CDC Human Influenza Virus Real-Time RT-PCR Diagnostic Panel (K190302). Performance characteristics of these extraction platforms with 2019-nCoV (SARS CoV-2) have not been demonstrated.

² CDC has confirmed that the external lysis buffer used with this extraction method is effective for inactivation of SARS-CoV-2.

³ CDC has compared the concentration of inactivating agent in the lysis buffer used with this extraction method and has determined the concentration to be within the range of concentrations found effective in inactivation of SARS-CoV-2.