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Tissue Culture (tc) PBS

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[1]FDA

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GenomeTrakr
Tech. support email: **genomeTrakr@fda.hhs.gov**

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This method was developed at the FDA's Center for Food Safety and Applied Nutrition for GenomeTrakr's pandemic response project, monitoring SARS-CoV-2 variants in wastewater. Protocols developed for this project cover wastewater collection, concentration, RNA extraction, RT-qPCR detection, library prep, genome sequencing, quality control checks, and data submission to NCBI. This method provides a reagent formula required in the rapid concentration of intact viruses from wastewater using a combination of PEG precipitation and ultracentrifugation.

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protocol ,

Sep 15, 2021

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Sep 15, 2021 Ruth Timme US Food and Drug Administration

Sep 23, 2021 Jessica Jones US Food and Drug Administration

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In steps of

Virus Concentration from Wastewater Using PEG Precipitation and Ultracentrifugation
Virus Concentration from Wastewater Using PEG Precipitation and Ultracentrifugation

Equipment:
Scientific Accumet XL 500 pH meter (Fisher Scientific Cat # 16-636-XL500A, or similar)

Reagents:
Sodium Chloride (NaCl) **Sigma**
Aldrich Catalog #S3014 Step 1.1 , or equivalent
Potassium Chloride **Sigma**
Aldrich Catalog #P9541 Step 1.2 , or equivalent
Potassium Phosphate dibasic (KH2PO4) **Sigma**
Aldrich Catalog #P9791 Step 1.3 , or equivalent
Sodium Phosphate dibasic anhydrous (Na2HPO4) **Sigma**
Aldrich Catalog #S5011 Step 1.4
, or equivalent

1X PBS

1 Dissolve components in deionized or ultrapure water to 1L.

1.1 Sodium Chloride (NaCl) **Sigma**
8.0 g **Aldrich Catalog #S3014**

1.2 Potassium Chloride **Sigma**
0.2 g **Aldrich Catalog #P9541**

1.3 **0.12 g**
Potassium Phosphate dibasic (KH2PO4) **Sigma**
Aldrich Catalog #P9791

1.4 **0.91 g**
Sodium Phosphate dibasic anhydrous (Na2HPO4) **Sigma**
Aldrich Catalog #S5011

2 Adjust pH **7.5**

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3 Autoclave 121 °C 00:15:00

15m

4 Store at 2-8 °C

For 2X tc PBS, double components in 1.1 - 1.4

10X tc PBS

5

Alternatively, 1X and 2X PBS can be made by dilution of
10X PBS **Sigma Aldrich Catalog #P5493** into sterile deionized or ultrapure water.

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