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Viral purification from faecal sample

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sarah.schulz^{1,2}

¹EMBL; ²NFDI4Microbiota

NFDI4Microbiota



Sarah Schulz

EMBL

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Protocol status: Working

We use this protocol and it's working

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Abstract

Protocol for the purification of viral particles from a faecal sample

Materials

SM Buffer, 0.2 µm syringe filter, 100-kDa-molecular-mass Amicon Ultra-15 Centrifugal Filter, DNase I , RNase



- 1 Homogenise 200 mg of faecal sample in 30 ml of SM buffer (50 mM Tris-HCl pH 7.5, 100 mM NaCl, 8 mM MgSO₄)
- 2 Centrifuge sample at 4000 rpm for 20 min, then filter through a 0.2 µm filter
- 3 Add filtrate to a 100-kDa-molecular-mass Amicon Ultra-15 Centrifugal Filter
- 4 Centrifuge filter tube at 4000 rpm for 3 min to concentrate sample
- 5 Repeat centrifugation until the entire diluted filtrate has been passed through the filter tube
- 6 Dilute filtrate with 30 ml SM buffer
- 7 Add diluted filtrate to a fresh 100-kDa-molecular-mass Amicon Ultra-15 Centrifugal Filter
- 8 Repeat centrifugation until a filtrate volume of at least 500 µl is reached
- 9 Add 10 U of DNase I and 10 U of RNase to the filtrate and incubate at 37°C for 30 min
- 10 Incubate samples at 65°C for 15 min to inactivate enzymes
- 11 Store at 4°C