

Figure 1

HEV-infected cell lysate (~1L)

Clarify/Filter thru 0.45um filter

Initial Crude HEV

TFF (300kD membrane)

TFF Retentate (~50 mL)

Spin 85K xg, 1.5h, 5C

Pellet

Suspend in PBS (~20mL)

Clarify/Filter thru 0.22um filter

UF to 2 mL (100kD membrane)

Filter sterilize

TFF HEV (~2 mL)

Supernatant (~50mL)

0.5% Triton X-100 + 0.1% LDS

Spin through 30% sucrose cushion 25K rpm, 8 h, 5C

Save pellet & suspend in PBS (~10 mL)

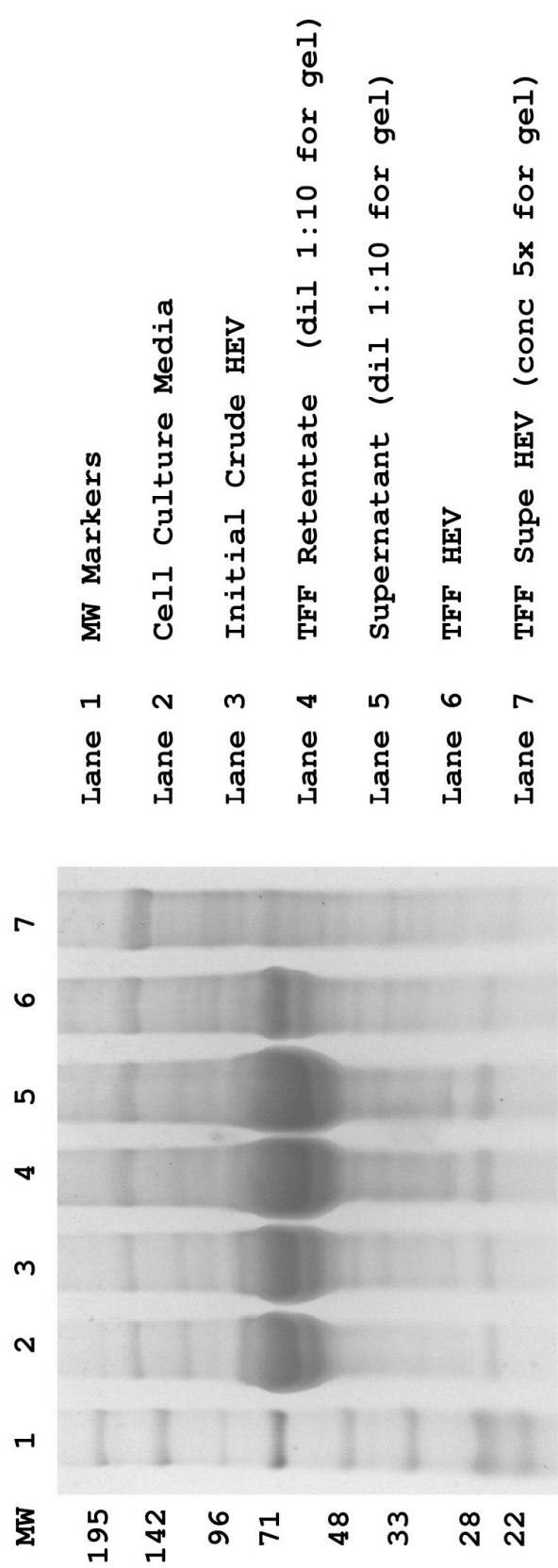
Clarify/Filter thru 0.22um filter

UF to 1-2 mL (100kD membrane)

Filter sterilize

TFF Supe HEV (~1-2 mL)

Figure 2



Parameter	Initial Crude	TFF Retentate	Supernatant	TFF HEV	TFF Supe HEV
Volume (mL)	500	50	50	1	1
A280	4	41	41	3.8	0.8
Log ₁₀ HEV RNA copies/mL	7.7	8.9	8.7	9.1	8.9

Figure 3

HEV-infected lysate	HEV-infected lysate	HEV-infected cell lysate
Clarify/Filter thru 0.45um filter	Clarify/Filter thru 0.45um filter	Clarify/Filter thru 0.45um filter
Initial Crude HEV	Initial Crude HEV	Initial Crude HEV
TFF (300kD membrane)	TFF (300kD membrane)	TFF (300kD membrane)
TFF Retentate	TFF Retentate	TFF Retentate
Spin 85xg, 1.5h, 5C	0.5% Triton Spin 85xg, 1.5h, 5C	0.5% Triton+0.1% LDS Spin 85xg, 1.5h, 5C
Pellet	Pellet	Pellet
Suspend in PBS	Suspend in PBS	Suspend in PBS
Clarify/Filter thru 0.22um filter	Clarify/Filter thru 0.22um filter	Clarify/Filter thru 0.22um filter
UF (100kD membrane)	UF (100kD membrane)	UF (100kD membrane)
Filter sterilize	Filter sterilize	Filter sterilize
TFF HEV	Triton TFF HEV	Triton/LDS TFF HEV

Figure 4

5'-CGGCTATCGGCCAGAAGTT-3' (SEQ ID NO: 1)
5'-CCGTGGCTATAACTGTGGTCT-3' (SEQ ID NO: 2)
5'-FAM-TTTTTACGC-ZEN-AGGCTGCCAAGGCC-3IABkFQ-3' (SEQ ID NO: 3)