

Materials List for:

Amplification of Near Full-length HIV-1 Proviruses for Next-Generation Sequencing

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Materials

Name	Company	Catalog Number	Comments
UltraPure 1 M Tris-HCl, pH 8.0	Invitrogen	15568025	Dilute to 5 mM for nested PCR
Nonidet P 40 Substitute solution	Sigma	98379	
Tween-20	Sigma	P9416	
Proteinase K Solution (20 mg/mL)	Promega	AM2548	
96 well thermocycler			Any 96 well thermocycler can be used
Platinum Taq DNA Polymerase High Fidelity	Invitrogen	11304011	
10x High Fidelity Buffer [600 mM Tris-SO ₄ (pH 8.9), 180 mM (NH ₄) ₂ SO ₄]	Invitrogen	11304011	
50 mM MgSO ₄	Invitrogen	11304011	
PCR Nucleotide Mix, 10 mM	Promega	C1141	
Ultrapure H ₂ O	Invitrogen	10977023	
PCR1 and PCR2 Primers			Desalted. Dilute to (100 μM) with H ₂ O
PCR Plate 96 Well, Half Skirt, Single Notch Corner, Clear	Axygen	PCR-96M2-HS-C	
Microseal 'B' Adhesive Seals	BioRad	MSB1001	Required to seal 96 well PCR plates
HIV-1 NL4-3 Infectious Molecular Clone (pNL4-3)	The following reagent was obtained through the NIH AIDS Reagent Program, Division of AIDS, NIAID, NIH: HIV-1 NL4-3 Infectious Molecular Clone (pNL4-3) from Dr. Malcolm Martin (Cat# 114).		Diluted to 10 ⁵ copies/mL and used as positive control
PCR plate spinner or benchtop centrifuge			
E-GEL 48 1% Agarose	Invitrogen	G800801	Precast 1% agarose gels (two gels used per 96 well plate). Any 1% agarose gel can be substituted. Contains ethidium bromide.
DirectLoad Wide Range DNA Marker	Sigma	D7058	Any ladder with a range up to 10 kb can be substituted
Mother E-Base device	Invitrogen	EBM03	Required for running precast 48 well 1% agarose gels
Gel Doc EZ Gel Documentation System	BioRad	1708270	Any visualisation system for ethidium bromide containing agarose gels can be used

PlateMax Peelable Heat Sealing	Axygen	HS-200	Heat sealing film for long term storage
96 Well 0.8 mL Storage Plate	ThermoFisher Scientific	AB0765	0.8 mL 96 well plate required for magnetic bead based PCR purification
Agencourt AMPure XP (PCR purification kit)	Beckman Coulter	A63880	Magnetic bead based PCR purification kit. Other PCR purification kits can be substituted here (e.g. QIAquick PCR Purification Kit (Qiagen Cat#28106))
Ethyl alcohol, Pure. 200 proof, for molecular biology	Sigma	E7023	
Magnetic Stand-96	Invitrogen	AM10027	
Microplate shaker			Optional
Buffer EB	Qiagen	19086	Elution buffer
Quant-iT PicoGreen dsDNA Assay Kit	Invitrogen	P11496	A fluorescence based stain for measuring dsDNA concentration
Microplate reader			
Nextera XT DNA Library Preparation Kit	Illumina	FC-131-1096	
Nextera XT Index Kit	Illumina	FC-131-2001	
Hard-Shell 96-Well PCR Plates	Biorad	HSP9601	Required for Nextera XT DNA library preparation kit
Library Quantification Kit - Illumina/ Universal	Kapa Biosystems	KK4824	Other library quantification kits can be substituted (e.g. JetSeq DNA Library Quantification Lo-Rox Kit (Bioline Cat#BIO-68029))
2100 Bioanalyzer	Agilent Technology		Automated electrophoresis system . Use in conjunction with a High Sensistivity DNA kit
High Sensistivty DNA kit	Agilent Technology	5067-4626	
Illumina MiSeq Platform	Illumina		