

Materials List for:

# Combining Analysis of DNA in a Crude Virion Extraction with the Analysis of RNA from Infected Leaves to Discover New Virus Genomes

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## Materials

Name	Company	Catalog Number	Comments
NaH <sub>2</sub> PO <sub>4</sub>	Sigma-Aldrich St. Louis MO	S5976	Grinding buffer for virus purification
Na <sub>2</sub> HPO <sub>4</sub>	Sigma-Aldrich	S0751	Grinding buffer for virus purification
Na <sub>2</sub> SO <sub>3</sub>	Thermo-Fisher Waltham, MA	28790	Grinding buffer for virus purification
urea	Thermo-Fisher	PB169-212	Homogenate extraction
Triton X-100	Sigma-Aldrich	X-100	Homogenate extraction
Cheesecloth	VWR Radnor, PA	21910-107	Filter homogenate
Tris	Thermo-Fisher	BP152-5	Pellet resuspension & DNA resuspension buffers
MgCl <sub>2</sub>	Spectrum, Gardena, CA	M1035	Pellet resuspension buffer
EDTA	Spectrum	E1045	Stops enzyme reactions
Proteinase K	Thermo-Fisher	25530	DNA resuspension buffer
phenol:chloroform:isoamylalcohol	Sigma-Aldrich	P2069	Dissolve virion proteins
DNase I	Promega	M6101	Degrade cellular DNA from extracts
95% ethanol	Sigma-Aldrich	6B-100	Virus DNA precipitation
Laboratory blender	VWR	58984-030	Grind leaf samples
Floor model ultracentrifuge & Ti70 rotor	Beckman Coulter, Irving TX	A94471	Separation of cellular extracts
Floor model centrifuge and JA-14 rotor	Beckman Coulter	369001	Separation of cellular extracts
Magnetic stir plate	VWR	75876-022	Mixing urea into samples overnight
Rubber policeman	VWR	470104-462	Dissolve virus pellet
2100 bioanalyzer Instrument	Agilent Genomics, Santa Clare, CA	G2939BA	Sensitive detection of DNA and RNA quality and quantity
2100 Bioanalyzer RNA-Picochip		5067-1513	Microfluidics chip used to move, stain and measure RNA quality in a 2100 Bioanalyzer
2100 Bioanalyzer DNA-High Sensitive chip		5067-4626	Microfluidics chip used to move, stain and measure DNA quality in a 2100 Bioanalyzer
Nanodrop spectrophotometer	Thermo-Fisher	ND-2000	Analysis of DNA/RNA quality at intermediate steps of procedures
Plant total RNA isolation kit	Sigma-Aldrich	STRN50-1KT	Isolate RNA for RNA-seq
RNase-free water	VWR	10128-514	Resuspension of DNA and RNA for NGS

RNA concentrator spin column	Zymo Research, Irvine, CA	R1013	Prepare RNA for RNA-seq
rRNA removal kit	Illumina, San Diego, CA	MRZPL116	Prepare RNA for RNA-seq
DynaMag-2 Magnet	ThermoFisher	12321D	Prepare RNA for RNA-seq
RNA enrichment system	Roche	7277300001	Prepare RNA for RNA-seq
Agarose	Thermo-Fisher	16500100	Gel analysis of DNA/RNA quality at intermediate steps of procedures
Ethidium bromide	Thermo-Fisher	15585011	Agarose gel staining
pGEM-T +JM109 competent cells	Promega, Madison, WI	A3610	Clone genome fragments
pFU Taq polymerase	Promega	M7741	PCR amplify virus genome
dNTPs	Promega	U1511	PCR amplify virus genome
PCR oligonucleotides	IDT, Coralvill, IA	Custom order	PCR amplify virus genome
Miniprep DNA purification kit	Promega	A1330	Plasmid DNA purification prior to sequencing
PCR clean-up kit	Promega	A9281	Prepare PCR products for cloning
pDRAW32 software	ACAClone		Computer analysis of circular DNA and motifs
MEGA6.0 software	MEGA		Molecular evolutionary genetics analysis
Primer 3.0	Simgene.com		
Quant-iT™ RiboGreen™ RNA Assay Kit	Thermo-Fisher	R11490	Fluorometric determination of RNA quantity
GS Junior™ pyrosequencing System	Roche	5526337001	Sequencing platform
GS Junior Titanium EmPCR Kit (Lib-A)	Roche	5996520001	Reagents for emulsion PCR
GS Jr EmPCR Bead Recovery Reagents	Roche	5996490001	Reagents for emulsion PCR
GS Junior EmPCR Reagents (Lib-A)	Roche	5996538001	Reagents for emulsion PCR
GS Jr EmPCR Oil & Breaking Kit	Roche	5996511001	Reagents for emulsion PCR
GS Jr Titanium Sequencing kit*	Roche	5996554001	Includes sequencing reagents, enzymes, buffers, and packing beads
GS Jr. Titanium Picotiter Plate Kit	Roche	5996619001	Sequencing plate with associated reagents and gaskets
IKA Turrax mixer		3646000	Special mixer used with Turrax Tubes
IKA Turrax Tube (specialized mixer)		20003213	Specialized mixing tubes with internal rotor for creating emulsions
GS Nebulizers Kit	Roche	5160570001	Nucleic acid size fractionator for use during library preparations
GS Junior emPCR Bead Counter	Roche	05 996 635 001	Library bead counter
GS Junior Bead Deposition Device	Roche	05 996 473 001	Holder for Picotiter plate during centrifugation
Counterweight & Adaptor for the Bead Deposition Devices	Roche	05 889 103 001	Used to balance deposition device with picotiter plate centrifugation
GS Junior Software	Roche	05 996 643 001	Software suite for controlling the instrument, collecting and analyzing data
GS Junior Sequencer Control v. 3.0	Roche		(Included in item 05 996 643 001 above)
GS Run Processor v. 3.0	Roche		(Included in item 05 996 643 001 above)

GS De Novo Assembler v. 3.0	Roche		(Included in item 05 996 643 001 above)
GS Reference Mapper v. 3.0	Roche		(Included in item 05 996 643 001 above)
GS Amplicon Variant Analyzer v. 3.0	Roche		(Included in item 05 996 643 001 above)