

GeoMx—NGS RNA Manual Equipment, Materials, & Reagents

Slide Prep

The following equipment ([see Table 1](#)), materials ([see Table 2](#)), and reagents ([see Table 3](#)) are recommended for this protocol but are **not supplied by NanoString**.

EQUIPMENT

Table 1: Equipment not provided by NanoString.

Equipment	Source	Part Numbers
Baking oven	Quincy Lab, Inc. (or comparable)	Various GC models
Hybridization oven including hybridization chamber *: Boekel Scientific RapidFISH Slide Hybridizer	Boekel Scientific	240200 for 120V or 240200-2 for 230V
or HybEZ II Hybridization System	ACDBio	
<ul style="list-style-type: none"> HybEZ oven Humidity control tray EZ-Batch Wash Tray EZ-Batch Slide Holder HybEZ Humidifying Paper (2 sheets) 		321710/321720 310012 321717 321716 310025
Water bath (up to at least 37°C)	Various	Various
UV Light Box (optional)	VWR	Various
5-quart Steamer	Nesco or Hamilton Beach	ST-25F 37530Z
	Note: Products from other vendors may require testing and optimization.	

* These hybridization ovens are designed to keep the slides hydrated and maintain a precise temperature overnight. NanoString does not recommend the use of any other hybridization ovens for GeoMx RNA DSP. Please contact NanoString with any concerns.

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MATERIALS

Table 2: Materials not provided by NanoString.

Materials	Source	Part Numbers
Pipettes for 5–1,000 µL	Various	Various
12-channel P20 multi-channel pipette	Various	Various
Filter Tips (RNase/DNase free)	Various	Various
Microtubes	Sarstedt (or comparable)	72.785.005
Superfrost Plus microscope slides	Fisher Scientific (or comparable)	12-550-15
Tissue Tek Slide Stain Dish (plastic Coplin Jars) or equivalent (recommend ~16)	Sakura (or comparable)	25608-904 or 25608-906
Instant-read digital thermometer	Various	Various
Humidity Chamber	Simport	M920-1
Benchtop protector sheet (fits inside the hybridization oven, optional)	Fisher Scientific (or comparable)	15235101
HybriSlip hybridization covers (22 mm x 40 mm x 0.25 mm) Note: this has been validated by NanoString. Do not use other products	Grace Bio-Labs	714022
RNase AWAY	ThermoFisher	7003PK
Heat/cold protectant handling glove	Various	Various
Forceps (for slide handling)	Various	Various
Aluminum foil	Various	Various

REAGENTS

Table 3: Reagents not provided by NanoString.

Reagent	Source/Part Number	Storage
DEPC-treated water	ThermoFisher, AM9922 (or comparable)	RT
10X phosphate buffered saline pH 7.4 (PBS)	Sigma-Aldrich, SKU P5368-10PAK, P5368-5X10PAK (or comparable)	RT
10% neutral buffered formalin (NBF) 	EMS Diasum, Cat # 15740-04 (or comparable)	RT
100% deionized formamide 	ThermoFisher, AM9342 or VWR, VWRV0606 (or comparable)	4°C (bring to RT before opening)
20X SSC (DNase, RNase free)	Sigma-Aldrich, S6639	RT
Proteinase K 	Proteinase K from Ambion, 2546 , or Thermo Fisher, AM2548 Note: Use of Proteinase K from any other vendor will require optimization of incubation times and concentration.	-20°C
1X Tris-EDTA pH 9.0	eBioscience™ IHC Antigen Retrieval Solution - High pH, 00-4956-58 (10X - will need to be diluted with DEPC-treated water)	4°C
Tris base	Sigma-Aldrich, 10708976001 (or comparable)	RT
Glycine	Sigma-Aldrich, G7126 (or comparable)	RT
Citrisolv or Xylene 	Fisher Scientific, Cat # 04-355-121 Sigma Aldrich, SKU 183164-100ML or 183164-500ML (or comparable)	RT
100% ethanol (EtOH): ACS grade or better 	Various	RT
10% Tween 20	Teknova, T0710 (or comparable)	RT

NGS RNA Library Prep

The following equipment ([see Table 4](#)), materials ([see Table 5](#)), and reagents ([see Table 6](#)) are recommended for this protocol but are **not supplied by NanoString**.

Equipment

Table 4: Equipment not provided by NanoString

Equipment	Manufacturer	Part Numbers
Pre-PCR hood (or designated space)	Various	Various
ALPS 50 plate sealer (see Materials list for compatible heat-sealing foil)	Thermo Fisher	AB-1443A
Plate spinner/centrifuge with plate adapter	Various	Various
Library quantitation device - qubit or similar	Various	Various
Vortex	Various	Various
Picofuge	Various	Various
Heatblock or thermal cycler	Various, such as Bio-Rad C1000 deep-well model	Various, such as 1851197
Plate spinner/centrifuge (up to at least 2000 g)	Various	Various
Capillary Electrophoresis device (e.g., Bioanalyzer)	Agilent	G2939BA
Illumina NGS instrument	Illumina	Various

Materials

Table 5: Materials not provided by NanoString

Materials	Manufacturer	Part Numbers
96-well PCR plates (RNase/DNase free)	Fisher Scientific	BC2496
Permeable membranes (dry-down seal)	Sigma	A9224
96 well plate stickers (freezing and storing plates)	Various	Various
Heat-sealing foil seals	Fisher Scientific	AB-0559
Microtubes (RNase/DNase free)	Sarstedt (or comparable)	72.785.005
PCR strip tubes (12-tube or 8-strip, RNase/DNase free)	Various	Various
Two* 12-channel P20 multi-channel pipettes	Various	Various
Filter Tips (RNase/DNase free)	Various	Various
Cooling block (optional)	Various	Various
Magnetic stand (e.g., DynaMag-96)	Invitrogen	12331D
Surface decontaminant (i.e., RNase Away or 10% Bleach)	Various	Various
	ThermoFisher	7002
DNA LoBind tubes (for library storage before sequencing)	Various	Various

* separate pipettes are needed for pre- and post PCR processes.

Reagents

Table 6: Reagents not provided by NanoString

Reagents	Manufacturer	Part Numbers
100% ethanol 	Various	Various
Agencourt AMPure XP	Beckman Coulter	A63880
Elution buffer (Tris-HCl 10 mM with 0.05% Tween-20, pH 8.0) *	Teknova	T1485
Bioanalyzer DNA High Sensitivity Kit	Agilent	5067-4626
PCR grade water	Various	Various

* Elution buffer can also be made by adding 5 mL 100mM Tris pH 8.0 and 2.5 mL 1% Tween-20 to 42.5 mL PCR-grade nuclease-free H₂O. Mix well.