

GeoMx— NGS RNA BOND RX 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
BOND RX ^m Compact, Fully Automated Advanced Stainer or BOND RX, Fully Automated Research Stainer	Leica Biosystems	Contact Leica Biosystems
BOND RX Controller, running BOND RX software version 6.0 and above	Leica Biosystems	Contact Leica Biosystems
Baking oven	Quincy Lab, Inc. (or comparable)	Various GC models
Hybridization oven including hybridization chamber *: Boekel Scientific RapidFISH Slide Hybridizer or HybEZ II Hybridization System	Boekel Scientific	240200 for 120V or 240200-2 for 230V
<ul style="list-style-type: none"> • HybEZ oven • Humidity control tray • EZ-Batch Wash Tray • EZ-Batch Slide Holder • HybEZ Humidifying Paper (2 sheets) 	ACDBio	321710/321720 310012 321717 321716 310025
Water bath (up to at least 37°C)	Various	Various
UV Light Box (optional)	VWR	Various

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* 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.

MATERIALS




Table 2: Materials not provided by NanoString.

Materials	Source	Part Numbers
BOND Research Detection System (includes 6x 30 mL Open Containers)	Leica Biosystems	DS9455
BOND Titration Kit (includes 50 inserts)	Leica Biosystems	OPT9049
BOND Universal Covertiles	Leica Biosystems	S21.2001
BOND Open Containers 30 mL (if additional needed)	Leica Biosystems	OP309700
BOND Titration Container Inserts (if additional needed)	Leica Biosystems	OPT9719
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	Sakura (or comparable)	25608-904 or 25608-906
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

REAGENTS

The following reagents are recommended for the GeoMx DSP Leica Biosystems RNA workflow but are not supplied by NanoString. BOND reagents are produced by Leica Biosystems; refer to Leica Biosystems product literature for safety information on these reagents.

Table 3: Reagents not provided by NanoString.

Reagent	Source/Part Number	Storage
BOND Dewax Solution, 1 Liter	Leica Biosystems, AR9222	4°C or RT
BOND Epitope Retrieval 2, 1 Liter	Leica Biosystems, AR9640	4°C
BOND Wash Solution 10X Concentrate, 1 Liter	Leica Biosystems, AR9590	4°C
Reagent-grade ethanol	Various	RT
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 (recommended)  or Leica Biosystems Enzyme Pretreat Kit	Ambion, 2546 Leica Biosystems, AR9951 Note: Using proteinase K from any other supplier will require optimization of timing and concentration.	-20°C 4°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

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.