

# Sample Submission Guideline (Novogene AMEA)

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This sample submission guideline is for customers in the Asia Pacific and Middle East region (AMEA), excluding mainland China. For any questions, please contact your Novogene representative.

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- If you require extraction services that are not covered in this document, or have sample or library types that are not listed, please reach out to [marketing\\_amea@novogeneait.sg](mailto:marketing_amea@novogeneait.sg) or your local sales representative.
- For guidance on how to prepare DNA or RNA samples from various sources, please refer to our [Sample Preparation Guide](#).
- It is recommended to double the sample amount when feasible, in case library re-construction is needed.
- Note that the sample volume for QC (2µL - 5µL) is excluded from the Sample Submission Guidelines.

## 1. Genome Sequencing

- It is recommended to suspend DNA samples in Tris-EDTA (TE) buffer, elution buffer (EB) or Tris-Borate (TB) buffer.
- High Molecular Weight (HMW) DNA samples should be in EB buffer.

### 1.1 Human Whole Genome Sequencing (WGS)

Library Type	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity (NanoDrop™/ Agarose Gel)
Human whole genome library preparation	Genomic DNA	≥ 100 ng	≥ 20 µL	≥ 5 ng/µL	OD260/280 = 1.8-2.0; no degradation, no contamination
	FFPE* DNA	≥ 400 ng	≥ 20 µL	≥ 15 ng/µL	Fragments longer than 1,000 bp
Human PCR-free library	Genomic DNA	≥ 1.2 µg	≥ 20 µL	≥ 50 ng/µL	OD260/280 = 1.8-2.0; no degradation, no contamination

\* FFPE: Formalin-Fixed, Paraffin-Embedded

### 1.2 Whole Exome Sequencing (WES)

Library Type	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity (NanoDrop™/ Agarose Gel)
Human WES library	Genomic DNA	≥ 300 ng	≥ 20 µL	≥ 15 ng/µL	OD260/280 = 1.8-2.0; no degradation, no contamination; Main peak is above 3000bp
	FFPE* DNA	≥ 400 ng	≥ 20 µL	≥ 15 ng/µL	Fragments longer than 1000 bp
	cfDNA/ctDNA	≥ 40 ng	≥ 20 µL	≥ 1 ng/µL	Fragments of 170 bp or multiples, no genomic DNA contamination
Mouse WES library	Genomic DNA	≥ 300 ng	≥ 20 µL	≥ 15 ng/µL	OD260/280 = 1.8-2.0; no degradation, no contamination

\* FFPE: Formalin-Fixed, Paraffin-Embedded

### 1.3 Plant & Animal Whole Genome Sequencing

Library Type	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity (NanoDrop™/ Agarose Gel)
Plant & Animal whole genome library preparation	Genomic DNA	≥ 100 ng	≥ 20 µL	≥ 5 ng/µL	OD260/280 = 1.8-2.0; no degradation, no contamination
Plant & Animal PCR-free library	Genomic DNA	≥ 1.2 µg	≥ 20 µL	≥ 50 ng/µL	
GBS (Genotyping by sequencing) library	Genomic DNA	≥ 600 ng	≥ 20 µL	≥ 20 ng/µL	OD260/280 = 1.8-2.0; no degradation, no RNA contamination

### 1.4 Microbial Whole Genome Sequencing & Metagenomics

Library Type	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity (NanoDrop™/ Agarose Gel)
Bacteria / Fungi whole genome library	Genomic DNA	≥ 100 ng	≥ 20 µL	≥ 5 ng/µL	OD260/280 = 1.8-2.0; no degradation, no contamination
Bacteria / Fungi PCR-free library	Genomic DNA	≥ 1.2 µg	≥ 20 µL	≥ 50 ng/µL	
Shotgun-based metagenomics library	Total DNA	≥100 ng	≥ 20 µL	≥ 5 ng/µL	OD260/280 = 1.8-2.0; no contamination; Main peak is above 500bp
Shotgun-based metagenomics PCR-free library	Total DNA	≥ 1.2 µg	≥ 20 µL	≥ 50 ng/µL	
Amplicon-based metagenomics	Total DNA	≥ 200 ng	≥ 20 µL	≥ 10 ng/µL	OD260/280 = 1.8-2.0; no degradation, no contamination

## 1.5 PacBio DNA Sequencing

Library Type	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity
PacBio DNA HiFi library	HMW* Genomic DNA (Human/ Plant/ Animal)	≥ 5.5 µg (Additional 5 ug per sample per Cell)	≥ 50 µL	≥ 70 ng/µL	OD260/280=1.75~2.0; OD260/230=1.5~2.6; NC/QC**=1.00~2.20; Fragments should be ≥30K
PacBio human low-input DNA HiFi library	HMW* Genomic DNA (Human)	≥ 1.2 µg (Only for 1 time sequencing in 1 Cell)	≥40 µL	≥30 ng/µL	OD260/280=1.75~2.0; OD260/230=1.5~2.6; NC/QC**=0.95~3.00; Fragments should be ≥30K
PacBio DNA HiFi library	HMW* Genomic DNA (Fungus/ Metagenomics)	≥ 5.5 µg (Additional 5 ug per sample per Cell)	≥ 50 µL	≥ 70 ng/µL	OD260/280=1.75~2.0; OD260/230=1.3~2.6; NC/QC**=1.00~2.20; Fragments should be ≥20K
PacBio DNA CLR library	HWM* Genomic DNA (Bacteria/ Fungus)	≥ 2 µg	≥40 µL	≥ 50 ng/µL	OD260/280=1.7~2.2; OD260/230=1.3~2.6; NC/QC**=0.95~3.00; Fragments should be ≥15K
PacBio Full-length 16S library	Genomic DNA	≥ 300 ng	≥30 µL	≥ 10 ng/µL	Clear main band; no degradation; no contamination

\* HMW: High Molecular Weight.

\*\* NC/QC: NanoDrop concentration/Qubit concentration

## 1.6 Nanopore Sequencing

Library Type	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity
Nanopore PromethION DNA library	HWM* Genomic DNA (Human/ Plant/ Animal)	≥ 8.5 µg	≥ 50 µL	≥ 100 ng/µL	OD260/280=1.75~2.0; OD260/230=1.4~2.6; NC/QC**=0.95~3.00 Fragments should be ≥30K
	HWM* Genomic DNA (Bacteria & Fungus)	≥ 6.5 µg	≥ 50 µL	≥ 60 ng/µL	OD260/280=1.7~2.2; OD260/230=1.3~2.6; NC/QC**=0.95~3.00 Fragments should be ≥15K
	HWM* Genomic DNA (Metagenomics)	≥ 5.5 µg	≥ 50 µL	≥ 80 ng/µL	OD260/280=1.7~2.5; OD260/230=1.1~2.6; NC/QC**=0.95~4.00 Fragments should be ≥10K

\* HMW: High Molecular Weight.

\*\* NC/QC: NanoDrop concentration/Qubit concentration

## 1.7 PCR Product Sequencing

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity (NanoDrop™/ Agarose Gel)
PCR-free library	PCR product	≥ 1.5 µg	≥ 20 µL	≥ 60 ng/µL	OD260/280 = 1.8-2.0; no degradation, no contamination
Library with PCR	PCR product	≥ 200 ng	≥ 20 µL	≥ 10 ng/µL	

For all Genome Sequencing products,

- It is recommended to double the sample amount when feasible, in case library re-construction is needed.
- Note that the sample volume for QC (2µL - 5µL) is excluded from the Sample Submission Guidelines.

## 2. RNA Sequencing

It is recommended to suspend RNA samples in RNase-free double-distilled water (ddH<sub>2</sub>O).

### 2.1 Transcriptome Sequencing

Library Type	Sample Type	Amount	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
Eukaryotic mRNA library (polyA enrichment)	Total RNA (animal/plant/fungus)	≥ 200 ng	≥ 20 µL	≥ 10 ng/µL	≥ 4.0, with flat baseline	OD260/280 ≥ 2.0; OD260/230 ≥ 2.0; no degradation, no contamination
	Total RNA (blood)	≥ 400 ng	≥ 20 µL	≥ 20 ng/µL	≥ 5.0, with flat baseline	
	Double stranded cDNA	≥ 100 ng	≥ 20 µL	≥ 5 ng/µL	Fragments between 400 bp-5000 bp, main peak at ~2000 bp	
Eukaryotic mRNA library (Globin removal & polyA enrichment)	Total RNA (human blood)	≥ 400 ng	≥ 20 µL	≥ 20 ng/µL	≥5.0, with flat baseline	
Eukaryotic strand-specific mRNA library (polyA enrichment)	Total RNA (animal/plant/fungus)	≥ 400 ng	≥ 20 µL	≥ 20 ng/µL	≥5.0, with flat baseline	
Eukaryotic strand-specific mRNA library (Globin removal & polyA enrichment)	Total RNA (human/mouse blood)*	≥ 400 ng	≥ 20µL	≥ 20 ng/µL	≥5.0, with flat baseline	
Prokaryotic strand-specific RNA library (rRNA depletion)	Total RNA	≥ 500 ng	≥ 20 µL	≥ 25 ng/µL	≥6.0, with flat baseline	
Dual RNA library (double rRNA depletion)	Total RNA	≥ 800 ng	≥20 µL	≥ 25 ng/µL	≥6.5, with flat baseline	
Metatranscriptome library (double rRNA depletion)	Total RNA	≥ 500 ng	≥ 20 µL	≥ 25 ng/µL	≥5.8, with flat baseline	
FFPE RNA library (TruSeq RNA Exome Panel)	FFPE total RNA	≥ 200 ng	≥ 15 µL	≥ 10 ng/µL	human only	

\* Globin clear applies to human blood only, Ribo-Zero Globin applies to human/mouse blood.

## 2.2 Eukaryotic Long Non-coding RNA Sequencing

Library Type	Sample Type	Amount	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
Directional RNA library (rRNA depletion)	Total RNA (animal/plant/fungus)	≥ 500 ng	≥ 20 µL	≥ 25ng/µL	≥ 5.5, with flat baseline	OD260/280 ≥ 2.0; OD260/230 ≥ 2.0; no degradation, no genomic contamination
	Exosome Total RNA	≥ 10 ng	≥ 10 µL	-	Fragments between 80-200nt, no peaks > 2000nt, FU* > 10	

\* FU: Fluorescent unit

## 2.3 Eukaryotic Small RNA Sequencing

Library Type	Sample Type	Amount	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
Eukaryotic small RNA library (18-40 bp insert)	Total RNA (animal)	≥ 2 µg	≥ 20 µL	≥ 50 ng/µL	≥ 7.5, with flat baseline	OD260/280 ≥ 2.0; OD260/230 ≥ 2.0; no degradation, no contamination
	Total RNA (plant/fungus)	≥ 2 µg	≥ 20 µL	≥ 50 ng/µL	≥ 7.0, with flat baseline	
	Exosome Total RNA	≥ 20 ng	≥ 20 µL	-	Fragments between 25-200nt	

## 2.4 Eukaryotic Circular RNA Sequencing

Library Type	Sample Type	Amount	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
Eukaryotic circRNA directional library (rRNA and linear RNA depletion)	Total RNA (animal)	≥ 2 µg	≥ 20 µL	≥ 50 ng/µL	≥ 7.0, with flat baseline	OD260/280 ≥ 2.0; OD260/230 ≥ 2.0; no degradation, no contamination
	Total RNA (plant)	≥ 2 µg	≥ 20 µL	≥ 50 ng/µL	≥ 6.5, with flat baseline	

## 2.5 Eukaryotic Whole Transcriptome Sequencing

Library Type	Sample Type	Amount	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
Eukaryotic lncRNA & small RNA library	Total RNA	≥ 2.5 µg	≥ 30 µL	≥ 50 ng/µL	≥ 7.5, with flat baseline (animal)	OD260/280 ≥ 2.0; OD260/230 ≥ 2.0; no degradation, no genomic contamination
Eukaryotic lncRNA & small RNA & circRNA library	Total RNA	≥ 4.5 µg	≥ 50 µL	≥ 50 ng/µL	≥ 7.0, with flat baseline (plant/fungus)	

## 2.6 Long Read Transcriptome Sequencing

Service	Sample Type	Amount**	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
PacBio Kinnex full-length RNA library/ Iso-Seq library	Total RNA	≥ 1.2 ug	≥ 30 µL	≥ 40 ng/µL	≥ 6.5 with flat baseline	OD260/280=1.8-2.2; OD260/230=1.3-2.5; NC/QC* ≤ 2

\* NC/QC: NanoDrop concentration/Qubit concentration

\*\* Highly recommend sending above 2ug if don't order per Cell.

## 2.7 Direct RNA Sequencing

Library Type	Sample Type	Amount	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
Nanopore direct RNA library	Total RNA (Animal)	≥ 20 µg	≥ 20 µL	≥ 10 ng/µL	≥ 6.5 with flat baseline	OD260/280=1.8-2.2; OD260/230=1.3-2.5; NC/QC* ≤ 2
	Total RNA (Plant / Fungus)	≥ 50 µg	≥ 20 µL	≥ 20 ng/µL	≥ 6.5 with flat baseline	OD260/280=1.8-2.2; OD260/230=1.3-2.5; NC/QC* ≤ 2

\* NC/QC: NanoDrop concentration/Qubit concentration

For all RNA Sequencing products,

- It is recommended to double the sample amount when feasible, in case library re-construction is needed.
- Note that the sample volume for QC (2µL - 5µL) is excluded from the Sample Submission Guidelines.

### 3. Epigenetics Sequencing

- It is recommended to suspend DNA samples in Tris-EDTA (TE) buffer, elution buffer (EB) or Tris-Borate (TB) buffer, and RNA samples in RNase-free double-distilled water (ddH<sub>2</sub>O).
- RIP-seq input controls should be rRNA-depleted prior to sample shipment.

Library Type	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity or fragment size (NanoDrop™/Agarose Gel)
Whole Genome Bisulfite Sequencing (WGBS)	Genomic DNA	≥ 200 ng	≥ 20 µL	≥ 5 ng/µL	0 < OD260/230 < 3; no degradation, no contamination
ChIP-seq	Enriched DNA	≥ 20 ng	≥ 20 µL	≥ 1 ng/µL	Main peak within 100 bp-500 bp
RIP-seq	Enriched RNA	≥ 80 ng	≥ 20 µL	≥ 3 ng/µL	Recommend fragments size 250-300bp

## 4. Premade Library Sequencing

Premade libraries should be colourless. Sub-libraries must be pooled together prior to library shipment.

### 4.1 Library Volume

Sequencing platform & sequencing strategy	Sequencing data amount	Volume requirements
NovaSeq X Plus 10B PE150	Lane sequencing	≥70 µL (additional 70 µL for one more lane)
NovaSeq X Plus 25B PE150	Lane sequencing	≥ 130 µL/lane (additional 130 µL for one more lane)
NovaSeq X Plus PE150	X < 30 G	≥ 15 µL
	30 G ≤ X < 100 G	≥ 30 µL
	100 G ≤ X < 375 G	≥ 70 µL
NovaSeq 6000 S4 PE150	X < 30 G	≥ 15 µL
	30 G ≤ X < 100 G	≥ 25 µL
	100 G ≤ X < 400 G	≥ 50 µL
	400 G ≤ X < 800 G	≥ 70 µL
	Lane sequencing	≥ 70 µL/lane (additional 70 µL for one more lane)
NovaSeq 6000 SP 500 Cycle/ SE50	X < 30 M reads	≥ 15 µL
	30 M ≤ X < 100 M reads	≥ 25 µL
	100 M ≤ X < 400 M reads	≥ 50 µL
	Lane sequencing	≥ 70 µL/lane (additional 70 µL for one more lane)
NovaSeq6000 SP PE50	Flowcell sequencing	≥ 140 µL/flowcell
DNB T7 PE150 (Illumina Linear Library)	X < 50 G reads	≥ 40 µL
	50 G ≤ X < 200 G reads	≥ 90 µL
	200 G ≤ X	≥ 100 µL
	Lane sequencing	≥ 50 µL/lane (additional 50 µL for one more lane)
DNB T7 PE150 (Illumina Cyclization Library)	Lane sequencing	≥ 120 µL/lane (additional 120 µL for one more lane)
DNB T7 PE100	Please check with our representatives	

## 4.2 Library Concentration

Sequencing platform & sequencing strategy	Library concentration
NovaSeq Platforms	≥ 2 ng/μL, quantified by Qubit® 2.0 (Life Technologies) or ≥ 2 nmol/L quantified by qPCR
DNB T7 PE150 (Illumina Linear Library)	≥ 2.5 ng/μL, quantified by Qubit® 2.0 (Life Technologies) or ≥ 12.6 nmol/L quantified by qPCR
DNB T7 PE150 (Illumina Cyclization Library)	≥ 1.5 ng/μL, quantified by Qubit® 2.0 (Life Technologies) or ≥ 7.5 nmol/L quantified by qPCR
DNB T7 PE100	Please check with our representatives

## 4.3 Library Size

- Library size includes: insert + adapters (120 bp) ± 50 bp (Does not apply to small RNA library)
- Libraries should have a single main peak, no multiple peaks, no adapter contamination, and no primer dimers.

Sequencing platform & sequencing strategy	Library size
NovaSeq PE150	> 300bp
DNBSEQ-T7 PE150	> 300bp
NovaSeq 6000 SE50/PE50	< 700bp
NovaSeq 500 cycles	> 370bp

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