Novogene | Advancing Genomics, Improving Life

NOVOGENE EUROPE SAMPLE REQUIREMENTS

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Contents

- Nucleic acid samples must be clear and completely colorless, exhibiting no visible tint or impurities.
- If you need extraction service from us, have any other sample types or library types not covered in this document, please contact us or your local sales representative. To extract nucleic acids from human tissue, please check the our guideline to handle human materials.
- If you need guidelines on how to prepare DNA or RNA samples from different sources, please check our Sample Preparation Guide.
- It is recommended to **double the sample amount** when possible.

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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)

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity (NanoDrop™/Agarose Gel)
Human WGS (350 bp insert size)	Genomic DNA	≥200 ng	≥20 μL	≥10 ng/μL	OD260/280 = 1.8-2.0; no degradation, no contamination
	FFPE* DNA	≥300 ng	≥30 μL	≥10 ng/μL	Fragments longer than 1000 bp
	cfDNA/ctDNA	≥30 ng	-	≥0.5 ng/μL	Fragments of 170 bp or multiples, no genomic DNA contamination
PCR-free human WGS	Genomic DNA	≥1.2 μg	≥20 μL	≥20 ng/μL	OD260/280 = 1.8-2.0; no degradation, no contamination

*FFPE: Formalin-Fixed, Paraffin-Embedded

1.2 WHOLE EXOME SEQUENCING (WES) / TARGET REGION SEQUENCING (TRS)

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity (NanoDrop™/Agarose Gel)
Human WES/TRS	Genomic DNA	≥300 ng	≥20 μL	≥15 ng/μL	OD260/280 = 1.8-2.0; no degradation, no contamination
	FFPE* DNA	≥500 ng	≥30 μL	≥15 ng/μL	Fragments longer than 1000 bp
	cfDNA/ctDNA	≥30 ng	-	≥0.5 ng/μL	Fragments of 170 bp or its multiples, no genomic DNA contamination
Mouse WES	Genomic DNA	≥300 ng	≥15 μL	≥20 ng/μL	OD260/280 = 1.8-2.0; no degradation, no contamination
	FFPE DNA	≥600 ng	≥30 μL	≥20 ng/μL	Fragments longer than 1000 bp

*FFPE: Formalin-Fixed, Paraffin-Embedded

1.3 PLANT & ANIMAL WHOLE GENOME SEQUENCING

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity (NanoDrop™/Agarose Gel)
Plant & Animal WGS (350 bp insert size)	Genomic DNA	≥200 ng	≥20 μL	≥10 ng/μL	OD260/280 = 1.8-2.0; no degradation, no contamination
PCR-free Plant & Animal WGS (350 bp insert size)	Genomic DNA	≥1.2 μg	≥20 μL	≥20 ng/μL	
PCR-free Plant & Animal WGS (custom size, ≤500 bp)	Genomic DNA	≥2 μg	≥20 μL	≥30 ng/μL	

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1.4 MICROBIAL WHOLE GENOME SEQUENCING & METAGENOMICS

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity (NanoDrop™/Agarose Gel)
Microbial WGS	Genomic DNA	≥200 ng	≥20 μL	≥10 ng/μL	OD260/280 = 1.8-2.0; no degradation, no contamination
Shotgun-based metagenomics	Total DNA	≥200 ng	≥20 μL	≥10 ng/μL	
PCR-free WGS / PCR-free shotgun-based metagenomics	Genomic DNA / total DNA	≥1.2 μg	≥20 μL	≥20 ng/μL	
Amplicon-based metagenomics	Total DNA	≥200 ng	≥20 μL	≥10 ng/μL	

1.5 PACBIO SEQUENCING

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity
PacBio DNA CLR library	HWM genomic DNA (Bacteria & Fungus)	≥1.5 μg	≥50 μL	≥70 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 DNA HiFi library	HWM genomic DNA (Plant & Animal)	≥5 μg	≥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
	HWM genomic DNA (Bacteria & Fungus)	≥5 μg	≥50 μL	≥70 ng/μL	OD260/280=1.75~2.2; OD260/230=1.3~2.6; NC/QC=1.00~2.20 Fragments should be ⩾20K
Pacbio Full -Length 16S/18S/ITS	Isolated DNA	≥300 ng	≥30 μL	≥10 ng/μL	OD260/280 = 1.8-2.0; no degradation, no contamination

*NC/QC = NanoDrop concentration/Qubit concentration

1.6 NANOPORE SEQUENCING

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity
Nanopore PromethION DNA library	HWM genomic DNA (Plant & Animal)	≥8 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 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 ⩾20K

*NC/QC = NanoDrop concentration/Qubit concentration

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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	

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2. RNA SEQUENCING

It is recommended to suspend RNA samples in RNAse-free double-distilled water (ddH_2O).

2.1 EUKARYOTIC MESSENGER RNA SEQUENCING

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
Eukaryotic mRNA (polyA enrichment)	Total RNA (animal)	≥200 ng	≥10 μL	≥20 ng/μL	≥4.0, with flat baseline	OD260/280≥2.0; OD260/230≥2.0; no degradation, no contamination
	Total RNA (plant, fungus)	≥200 ng	≥10 μL	≥20 ng/μL	≥4.0, with flat baseline	
	Total RNA (blood)	≥400 ng	≥20 μL	≥20 ng/μL	≥5.0, with flat baseline	
	Double stranded cDNA	≥100 ng	≥10 μL	≥10 ng/μL	Fragments between 400 bp-5000 bp, main peak at ~2000 bp	
Strand-specific Eukaryotic mRNA (polyA enrichment)	Total RNA (animal)	≥400 ng	≥20 μL	≥20 ng/μL	≥5.0, with flat baseline	
	Total RNA (plant, fungus)	≥400 ng	≥20 μL	≥20 ng/μL	≥5.0, with flat baseline	
	Total RNA (blood)	≥400 ng	≥20 μL	≥20 ng/μL	≥5.0, with flat baseline	

2.2 TRANSCRIPTOME SEQUENCING

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
Eukaryotic strand-specific lncRNA (rRNA depletion)	Total RNA (animal)	≥500 ng	≥10 μL	≥50 ng/μL	≥5.5, with flat baseline	OD260/280≥2.0; OD260/230≥2.0; no degradation, no contamination
	Total RNA (plant, fungus)	≥500 ng	≥10 μL	≥50 ng/μL	≥5.5, with flat baseline	
	Total RNA (blood)	≥500 ng	≥10 μL	≥50 ng/μL	≥5.5, with flat baseline	
	Exosome RNA (human, mouse)	≥5 ng	≥10 μL	-	Fragments between 25-200nt, FU*>10	
Prokaryotic strand-specific RNA (rRNA depletion)	Total RNA	≥500 ng	≥10 μL	≥50 ng/μL	≥6.0, with flat baseline	
Dual RNA (double rRNA depletion)	Total RNA	≥800 ng	≥20 μL	≥40 ng/μL	≥6.5, with flat baseline	
Metatranscriptome (double rRNA depletion)	Total RNA	≥800 ng	≥20 μL	≥40 ng/μL	≥5.8, with flat baseline	

*FU = Fluorescent unit

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2.3 EUKARYOTIC SMALL RNA SEQUENCING

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
Eukaryotic sRNA (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 RNA	≥10 ng	≥10 μL	-	Fragments between 25-200nt, FU*>10	

*FU = Fluorescent unit

2.4 EUKARYOTIC CIRCULAR RNA SEQUENCING

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
Eukaryotic circRNA (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, fungus)	≥2 μg	≥20 μL	≥50 ng/μL	≥6.5, with flat baseline	

2.5 EUKARYOTIC WHOLE TRANSCRIPTOME SEQUENCING

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

2.6 LONG READ TRANSCRIPTOME SEQUENCING

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	RIN (Agilent 2100)	Purity (NanoDrop™)
Pacbio Iso-seq (polyA enrichment)	Total RNA	≥600 ng	≥15 μL	≥40 ng/μL	≥6.5 with flat baseline	OD260/280=1.8-2.2; OD260/230=1.3-2.5; NC/QC*≤2
Nanopore RNA (polyA enrichment)	Total RNA	≥100 ng	≥10 μL	≥10 ng/μL		

*NC/QC = NanoDrop concentration/Qubit concentration

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3. EPIGENETICS SEQUENCING

It is recommended to suspend RNA samples in RNAse-free double-distilled water (ddH2O), and DNA samples in Tris-EDTA (TE) buffer, elution buffer (EB) or Tris-Borate (TB) buffer.

RIP-seq input controls should be rRNA-depleted prior to sample shipment.

Service	Sample Type	Amount (Qubit®)	Volume	Concentration	Purity or fragment size (NanoDrop™/Agarose Gel)
Whole Genome Bisulfite Sequencing (WGBS)	Genomic DNA	≥100 ng	≥20 μL	≥5 ng/μL	0 < OD260/230 < 3; no degradation, no contamination
Reduced Representation Bisulfite Sequencing (RRBS)	Genomic DNA	≥800 ng	≥20 μL	≥40 ng/μL	0 < OD260/230 < 3; no degradation, no contamination
ChIP-seq	Enriched DNA	≥5 ng	≥10 μL	-	Main peak within 100 bp and 500 bp
RIP-seq	Enriched RNA	≥20 ng	≥10 μL	≥2 ng/μL	Fragments longer than 80 nt

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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 requirement
NovaSeq X Plus PE150	<30 Gb	≥15 μL
	30 Gb ≤ X < 100 Gb	≥30 μL
	100 Gb ≤ X < 375 Gb	≥70 μL
	375 Gb ≤ X < 1 Tb	≥130 μL
	Lane sequencing on 10B	≥70 μL (additional 70 μL for one more lane)
	Lane sequencing on 25B	≥130 μL (additional 130 μL for one more lane)
NovaSeq 6000 PE150	<30 Gb	≥15 μL
	30 Gb ≤ X < 100 Gb	≥25 μL
	100 Gb ≤ X < 400 Gb	≥50 μL
	400 Gb ≤ X ≤ 800 Gb	≥70 μL
	Lane sequencing	≥70 μL (additional 70 μL for one more lane)
NovaSeq 6000 SE50/PE50/PE250	<20 M reads	≥15 μL
	20 M reads ≤ X < 50 M reads	≥25 μL
	50 M reads ≤ X ≤ 150 M reads	≥50 μL
	Lane sequencing	≥70 μL (additional 70 μL for one more lane)

For flow cell sequencing projects where custom sequencing primers (R1, R2, index) are needed, please send them with 100 μM concentration and 50 μL volume.

4.2 LIBRARY CONCENTRATION

- ≥ 2 ng/μL, quantified by Qubit® 2.0 (Life Technologies)
  ≥2 nM, quantified by qPCR

4.3 LIBRARY SIZE

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

Sequencing platform & sequencing strategy	Library size
NovaSeq 6000 SE50/PE50	<700 bp
NovaSeq 6000 PE150	300 bp ~ 600 bp
NovaSeq 6000 PE250	>370 bp