

# NGS Sample Preparation Guide

## DNA Quality

- Double-strand, non degraded, and containing no particulate matter
- Purified by column or gel purification protocols
- A260/280 ratio of 1.8 or above

## RNA Quality

- RNA integrity Number value should be greater than 8
- 28S rRNA band at 4.9kb should be twice the integrity of the 18S rRNA band at 1.9kb

## Sample Quantity

- DNA sample should be measured by “Picogreen method”.
- RNA sample should be measured by Ribogreen or 2100 bioanalyzer RNA chip.

GS-FLX



	Sample Type	Conc.(ng/ $\mu$ l)	Vol.( $\mu$ l)	Total Amount
Shotgun library (Rapid Shotgun)	High Molecular Weight DNA( >1.5kb)	>50ng/ $\mu$ l	>10 $\mu$ l	>500ng
	Low molecular weight DNA(70-500bp)	>10ng/ $\mu$ l	>10 $\mu$ l	>100ng
Mate Pair library	3kb insert	>100ng/ $\mu$ l	>50 $\mu$ l	>5 $\mu$ g
	8kb insert	>100ng/ $\mu$ l	>150 $\mu$ l	>15 $\mu$ g
	20kb insert	>100ng/ $\mu$ l	>300 $\mu$ l	>30 $\mu$ g
Rapid cDNA library	purified mRNA	>15ng/ $\mu$ l	>19 $\mu$ l	>200ng
	Total RNA	>1 $\mu$ g/ $\mu$ l	>50 $\mu$ l	>50 $\mu$ g




	Sample Type	Conc.(ng/ $\mu$ l)	vol.( $\mu$ l)	total amount
<b>Shotgun (TruSeq)</b>	DNA	>50ng/ $\mu$ l	>20 $\mu$ l	>1 $\mu$ g
<b>Mate Pair Library (up to 5kb)</b>	DNA	>100ng/ $\mu$ l	>100 $\mu$ l	>10 $\mu$ g
<b>mRNA Library</b>	total RNA	>20ng/ $\mu$ l	>50 $\mu$ l	1~10 $\mu$ g
	mRNA	-	-	>100ng
<b>small RNA Library</b>	total RNA	>200ng/ $\mu$ l	>5 $\mu$ l	1~10 $\mu$ g
	small RNA	-	>5 $\mu$ l	all small RNA from 1~10 $\mu$ g of total RNA
<b>ChIP seq library*</b>		>0.2ng/ $\mu$ l	>50 $\mu$ l	>10ng

\* 200~300bp size sonicated samples are desirable, please provide with control DNA. If you do not wish to sequence the control DNA please contact us in advance.



## Targeted Sequencing

	Sample Type	Conc.(ng/ $\mu$ l)	Vol.( $\mu$ l)	Total Amount
<b>Nimblegen EZ Exon</b>	DNA	>50ng/ $\mu$ l	>20 $\mu$ l	>1 $\mu$ g
<b>Nimblegen Array</b>	DNA	>50ng/ $\mu$ l	>60 $\mu$ l	>1 $\mu$ g
<b>Sureselect</b>	DNA	>50ng/ $\mu$ l	>60 $\mu$ l	>1 $\mu$ g
<b>TruSeq Exome Enrichment</b>	DNA	>50ng/ $\mu$ l	>20 $\mu$ l	>1 $\mu$ g

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# NGS Sample Submission

## 1. DNA Submission Condition

- Submit sample DNA in TE buffer at a minimum concentration of 100ng/ $\mu$ l in a clearly labeled 1.5~2.0ml microcentrifuge tube sealed with parafilm tightly.
- Include a brief description of the DNA extraction and/or enrichment protocol used in the “Comments” fields of the sample order sheet.
- Certain carriers and pellet paint types are not compatible with aspects of our sequencing technology. Non-robust results have been observed from samples where Lithium Chloride (LiCl), fluorescent pellet paint, or tRNA was used prior to precipitation and resuspension for sample preparation input.

## 2. RNA Submission Condition

- The best condition for RNA storage or delivery is Ethanol precipitation condition (stable for 1 year at -20C); All solutions should be used with DEPC-treated water.
  1. Add 0.1 volume of 3 M Sodium Acetate (NaAce, pH7-8) to RNA solution and mix gently.
  2. Add 2 volume of 100% ethanol to RNA solution and mix gently.ex) If you have 100 $\mu$ l of RNA solution, add 10 $\mu$ l of 3 M Sodium Acetate (NaAcetate, pH7-8) and 220 $\mu$ l of 100% ethanol to RNA solution and mix well gently.
  3. Send with Blue Ice Packs.
- Submit sample RNA in a clearly labeled 1.5~2.0ml microcentrifuge tube sealed with parafilm tightly.
- Include a brief description of the RNA extraction and/or enrichment protocol used in the “Comments” fields of the order sheet.
- Certain carriers and pellet paint types are not compatible with aspects of our sequencing technology. Non-robust results have been observed from samples where Lithium Chloride (LiCl), fluorescent pellet paint, or tRNA was used prior to precipitation and resuspension for sample preparation input.

## 3. Shipping Samples to Macrogen

- Place sealed individual microcentrifuge tubes in a 50ml disposable screw up tube for additional insulation during shipment. To prevent sample tubes from moving during shipment, pack any remaining space in the 50ml tube with clean tissue paper prior to sealing.
- You should include the followings in the package.
  1. Prepared Sample with ice pack or dry ice.
  2. Macrogen Sample Order Sheet (with QC files)
  3. Client Information

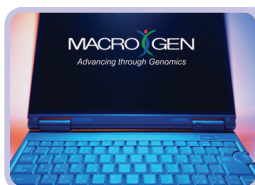
- Ship samples to :

**Macrogen Inc.**

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