



# **Sample Submission Guidelines for Sequencing Services**

**Kinghorn Centre for Clinical Genomics  
Sequencing Laboratory (KCCGSL)**

February 2022

# Instructions for submitting samples

Sequencing is performed by the Kinghorn Centre for Clinical Genomics Sequencing Laboratory (KCCGSL), a wholly owned and operated unit of the Garvan Institute of Medical Research located at 370 Victoria Street, Darlinghurst NSW 2010. To get started with your project, please follow the process below.

## Overview of sample submission process



1. Contact the customer support team to discuss your project via:
  - a. Email: [kccgseq@garvan.org.au](mailto:kccgseq@garvan.org.au)
  - b. In person: on level 7 at The Kinghorn Centre for Clinical Genomics Sequencing Laboratory
2. Receive a quote via email
3. Notify the KCCGSL team of quote acceptance by emailing confirmation and a purchase order (PO) PDF to [kccgseq@garvan.org.au](mailto:kccgseq@garvan.org.au)
4. Receive login details to the KCCGSL Customer Portal:  
<https://apps.kccg.garvan.org.au/sample-submission-x/#/home>
5. Receive email with link to manifest
6. KCCGSL-supplied FluidX barcoded tubes will be posted to your provided shipping address
7. Fill in and submit the manifest
8. Wait for 'Manifest Accepted' email
9. Aliquot your samples into the KCCGSL-supplied FluidX barcoded tubes
10. Submit samples to KCCGSL along with manifest label as per shipping requirements below (page 8)

## Sample preparation requirements

The input sample quality and quantity (DNA/RNA or library) are critical determinants for the success of a sequencing run and the subsequent generation of robust sequencing data.

Samples that do not pass the following requirements, as assessed using our in-house QC protocols, may continue to be processed (at risk) or removed from processing. Our team will provide information regarding such samples to the project lead/coordinator.

Given the nature of specific sample types e.g. FFPE tissue or submitted libraries, the laboratory cannot guarantee QC performance for library or sequencing results derived from these specimen types. To assist with the optimal performance of your samples, please notify our customer support team if any of the below submission requirements cannot be met for your samples, and/or if there are special requirements for the handling of your samples.

## Genomic DNA

- DNA should be extracted using Qiagen DNeasy/Tissue Extraction kits or equivalent
- Samples must be pure, intact, and of high molecular weight
- Samples must be free of contaminating nucleic acids
- Samples must not be amplified (PCR, WGA, MDA etc.)
- DNA should be submitted in Qiagen Elution Buffer (EB), Illumina Resuspension Buffer (RSB) or similar 10mM Tris/1 mM EDTA
- Sample concentration must be obtained using a fluorometric assay specific to dsDNA (e.g. Picogreen, Qubit BR assay)
- Sample quality to be assessed prior to submission, preferably using a Bioanalyzer, LabChip GX or similar
- Samples must meet the following specifications (QC Metrics):

Protocol	Vol. (uL)	Insert Size	A260:280	Total
WGS: KAPA Hyper PCR-Free Library Preparation Kit	100	400 bp	>=1.8	>=1.2 ug
WGS: Illumina DNA PCR-Free Prep Kit	80	450bp	>=1.8	>=1.2 ug
WGBS: ACCEL – NGS Methyl-Seq DNA Library Kit	100	350 bp	>=1.8	>=0.5 ug

## RNA (Total RNA, mRNA)

- Samples should be extracted using Qiagen RNeasy Extraction Kits or similar.
- RNA must be submitted in DNase/RNase free water
- Sample concentration must be obtained using a fluorometric assay specific to RNA (e.g. Qubit RNA assay)
- Sample quality must be assessed prior to submission, preferably using a Bioanalyzer, LabChip GX or similar
- Samples must meet the following specifications (QC Metrics):

Protocol	Vol. (uL)	Conc. (ng/uL)	RIN	A260:280	Total
KAPA RNA HyperPrep Kit with RiboErase* / KAPA mRNA HyperPrep Kit	20 - 30	50 - 100	$\geq 7.0$	$\geq 2.0$	$\geq 1.5\mu\text{g}$

\* Only compatible with Human, Mouse and Rat. RiboErase method can also be used for degraded RNA samples (RIN <7.0) or FFPE samples. Please consult with customer support team for low quality RNA samples.

## Submitted Libraries NovaSeq

### 6000 platform

The following table can be used as a guide for the submission requirements of pooled submitted libraries. Please contact the customer support team to discuss submitted library projects in detail before submitting to ensure optimal sequencing results are achieved for the project.

Pooled libraries (Standard Mode)	Minimum Vol. (uL)	Minimum Conc. (nM)
SP/S1 Flow cell (per flow cell)	130	4
S2 Flow cell (per flow cell)	200	4
S4 Flow cell (per flow cell)	350	4

Pooled libraries (XP Mode)	Min. Vol. (uL) for 1 lane	Min. Vol. (uL) for 2 lanes	Min. Vol. (uL) for 3 lanes	Min. Vol. (uL) for 4 lanes	Min. Conc. (nM)
SP/S1 Flow cell	35	60	NA	NA	4
S2 Flow cell	40	65	NA	NA	4
S4 Flow cell	50	85	120	150	4

## NextSeq 550

Pooled libraries	Minimum Vol. (uL)	Minimum Conc. (nM)
High Output Flow cell (per flow cell)	20	4
Mid Output Flow cell (per flow cell)	20	4

## Manifest and tubes

Customers must fill-in the online electronic manifest via the KCCGSL Customer Portal detailing sample characteristics. Customers must transfer samples into the provided FluidX 2D barcoded tubes and ensure the caps are sealed tightly.

- The manifest is pre-filled with the associated tube barcode(s), providing a unique identification code for each submitted sample. Please ensure that sample details for each barcode matches the sample aliquoted into that tube. In addition, ensure all the necessary details are filled out prior to submitting the manifest.
- KCCGSL will, within a reasonable time period of manifest receipt, review the manifest against the quote and either:
  - ✓ Approve the manifest
  - ✓ Decline the manifest and cancel the order; or
  - ✓ If the manifest is identified as non-conforming, KCCGSL will contact the submitting team/individual and seek agreement for KCCGSL to issue a revised quote (which may be subject to special conditions which will be specified).
- If KCCGSL accepts the manifest, we will notify the customer and request the samples be sent to KCCGSL.
- KCCGSL will, within a reasonable time period of sample receipt, inspect the samples in accordance with these guidelines and either:
  - ✓ Accept the samples
  - ✓ Decline to accept the samples and cancel the order
  - ✓ Decline to accept the samples and hold the order pending receipt of further information or further samples; or
  - ✓ If the samples are identified as non-conforming, contact the submitting team/individual to seek agreement for revised sample acceptance; this is subject to special conditions which may be specified in a revised quote (failing agreement, KCCGSL may decline to accept the samples and cancel the order).
- Samples sent in any container other than the FluidX 2D barcoded tubes provided by KCCGSL must be arranged prior to sending the samples. We cannot accept samples

until we have received and approved the corresponding manifest. You will receive an email notification advising you to prepare your samples for shipment.

## Disclaimer

KCCGSL rigorously tracks the identity of submitted samples throughout all stages of testing based on the FluidX tube 2D barcode, and information provided by the customer on the accompanying online manifest, to ensure that we always deliver the correct genomics data for the correct sample.

However, for the purposes of providing our services, since KCCGSL has no role in the primary collection and aliquoting of samples, it is assumed that the customer or submitter accepts full responsibility for the correct original identification of all samples submitted to us for testing.

This means that the customer is responsible for ensuring that the correct sample has been aliquoted into the correct tube (e.g. FluidX 2D Barcode Tubes or equivalent) and that this sample matches the sample identity as indicated on the online manifest entered on the KCCGSL submission portal.

## Turnaround time and delivery

The turnaround time (TAT), if any is specified in the quote, is inclusive and begins from the day of receipt and approval of samples in the laboratory until data delivery of all data files to the specified recipients. In the event any samples or projects don't meet agreed QC metrics, customers will be notified to seek an agreement on whether/how to proceed, and the subsequent turnaround time will be revised.

Application	Typical TAT for return of FASTQ (up to)
Whole Genome Sequencing	6 weeks
Submitted Libraries	6 weeks
RNA Sequencing (Transcriptome)	6 weeks <sup>1</sup>
Oncology Panel (TSO500)	10 weeks <sup>2</sup>
Whole Genome Bisulphite Sequencing	10 weeks
Additional Bioinformatics to FASTQ delivery	Consult with KCCGSL <sup>3</sup>

<sup>1</sup> TAT if the RNA-seq project occupies a full flow cell. Turnaround time may be extended for projects only partially occupying a flow cell (e.g. single lane)

<sup>2</sup> Up to 10 weeks for TSO500 oncology panel results pending sample batch size, also utilises complete flow cell <sup>3</sup> Up to an additional week required for each additional bioinformatics pipeline e.g. gVCF for WGS will require an additional week and gVCF + structural variants will require 2 weeks.

Faster turnaround times can be arranged under special circumstances and should be discussed with members of the KCCGSL team upon initial enquiry.

## Provision and retention of data files

- Data files generated from the samples submitted will be made available through a cloud-based platform. Additional delivery methods e.g. storage media devices can be arranged at additional cost.
- If the data files are to be made available to the customer on a cloud-based platform, the customer must authorise Garvan to provide the data files to the provider of that cloud-based platform.
- Garvan will retain the data files for a period of 90 days from the date of delivery (retention period). If the customer does not receive the data file or the data file is otherwise unavailable to the customer on the relevant data delivery platform, the customer is responsible for contacting Garvan within the retention period to request that the data file be delivered again. Garvan may not be able to provide data files after the retention period. Any data files provided after the retention period will be at an additional cost to the customer.
- Delivery of the data file is undertaken or facilitated by third-parties and Garvan is not liable to the customer for the performance or non-performance of any such third-party in respect to the delivery of, or failure to deliver, the data file.
- Where the customer selects the cloud-based platform option for delivery of the data file, Garvan will be required to disclose personal information relating to the customer's personnel to the cloud-based platform provider to enable delivery of the data file (for example, contact details necessary to create an account). The customer must ensure personnel consent to such personal information being provided.
- All personal information will be handled by the cloud-based platform in accordance with their privacy and online security policy. Please note, the Garvan has not taken steps to ensure the cloud-based provider doesn't breach the Australian Privacy Principles.
- **Data Delivery Platform** means a cloud-based platform for transfer and/or storage of a data file (or physical data storage media if specified in the quote).

- **Data File** means FASTQ format of DNA sequence data (or another available data file format agreed with the customer and specified in the quote).

## Sample retention

As per clause 12 of the Terms and Conditions unless otherwise agreed upon in a quote.

## Shipping requirements

- Please ensure that all barcoded tubes sit securely in the FluidX tube rack (if provided) and the lid is locked using the orange clips on the side. Ensure the rack containing the barcoded tubes is packaged in an appropriately rigid shipping container.
- If no FluidX tube rack is provided, prevent damage to your FluidX 2D tubes in transit by ensuring your tubes are stored securely as follows:
  - Place the primary receptacle (FluidX 2Dtube) into a secondary receptacle e.g. tube rack, Falcon tube or BioBottle
  - Place the secondary receptacle into a tertiary receptacle e.g. padded outer packaging
  - Package the tertiary receptacle into an appropriately rigid shipping container
- KCCGSL will provide customers with a shipping label (sent to customer with FluidX tubes) noting the project manifest ID. Please affix this label to the outside of the shipping container.
- DNA samples can be shipped on ice packs (domestic) or on dry ice (international).
- RNA samples must be shipped on dry ice.
- Submitted libraries must be shipped on dry ice.
- Attach the provided shipping label addressed to KCCGSL as per below:

ATTN:

KCCG Sequencing Laboratory

Garvan Institute of Medical Research

Precinct Loading Dock

West Street (off Burton Street)

Darlinghurst NSW 2010

Australia

Phone: +61 2 9355 5763

- At the time of shipping, please send an email notification to [kccgseq@garvan.org.au](mailto:kccgseq@garvan.org.au) with your express mail tracking number, as this will allow us to monitor your shipment.
- We will notify you when the samples have been received and approved (under these guidelines) in the laboratory.

### **For international customers**

In addition to the above instructions, please ensure that the following documents are clearly attached to the outer package:

- AQIS Import Permit (supplied by KCCGSL)
- Manufacturer's Declaration (supplied by KCCGSL)
- 4 copies of Customs Declaration (customer to provide)

We recommend that you state a value of less than AUD\$1,000 (or equivalent) on any paperwork you provide, so as to avoid triggering a tax invoice. Please note, this is not the value of the sequencing project.

We currently recommend World Courier, LabCabs, DGI and Logical Freight Solutions. We strongly advise against the use of DHL or FedEx for shipment of samples to Australia.