USER GUIDE



FORMALIN-FIXED PARAFFIN-EMBEDDED (FFPE) TISSUE





Today's Research. Tomorrow's Medicine.

ABOUT US

The GSC's CAP, DAP and ISO 27001 certified technology platform is a high-throughput, large-scale DNA and RNA sequencing and analysis facility that has been designed to maximize analytical capacity, diversity, efficiency, scalability and flexibility. Our state-of-the-art clinical, sequencing, bioinformatics and proteomics platforms are ready to be put to use for your research or clinic.

We partner with researchers, guide experimental design, execute high quality processing of complex and valuable biological samples and provide extensive bioinformatics analyses with the aim of making genomics research accessible to our partners and collaborators within the scientific community.

Please feel free to <u>contact us</u> if you have any questions about the services we provide.





www.bcgsc.ca | info@bcgsc.ca | @BCCancer_GSC





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ABOUT THIS GUIDE

This user guide is for the preparation and submission of **formalin-fixed paraffin-embedded (FFPE) tissue**. If you are submitting a different sample type, please refer to the other user guides on <u>our website</u>.







SAMPLE PREPARATION

SUBMITTING SAMPLES FOR TOTAL NUCLEIC ACID (TNA) EXTRACTION

We accept a variety of samples for TNA extraction and sequencing at the GSC including blood, fresh frozen tissues, formalin-fixed paraffin-embedded (FFPE) tissues, saliva and buccal (cheek) swabs. Cell samples are accepted in tube format. Non-archival (FFPE) tissue samples must be frozen at -80°C. Blood or saliva samples can be submitted at ambient temperature according to instructions on the collection tubes.

This user guide will focus on FFPE tissue submissions.

The following FFPE tissue starting material guidelines work well for human and mouse samples. If working with other organisms, please <u>contact us</u>.

SAMPLE TYPE	SUBMISSION REQUIREMENTS
FFPE Tissue	 All samples must be frozen at -80°C Samples must be submitted in clearly labeled 1.0 mL Matrix tubes (Thermo Scientific, <u>Cat#</u> 3740) *Tubes can be provided, if necessary
	 Scrolls: min 120 mm² x 10 µm (for tissue surface area ≤ 120 mm², up to 3 scrolls per 1.0 mL Matrix tube) Cores: 2.5 mm x 1-3 mm (up to 2 cores per Matrix tube)

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ONLINE SAMPLE SUBMISSION

The online submission form must be completed and approved prior to submitting your samples to the GSC.

Our "Online Submissions Webpage" can be found <u>here</u>. Please use Firefox or Google Chrome browsers.

This guide to online submissions will provide helpful information that will assist you in navigating our online submission website.

Please write to GSC_submissions@bcgsc.ca if you have any questions.





GETTING STARTED

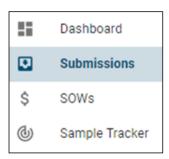
- 1. You can access the GSC Sample Submission Interface by entering the link below in Firefox or Google Chrome: https://www.bcgsc.ca/samplesubmissions
- 2. Enter your username and password, and then click Login:

GSC Sample Submission Interface



Forgot Username or Password?

3. To start a new submission, click Dashboard or Submissions on the left side panel:

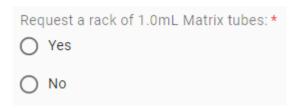


- Click the [Dashboard) icon on the left side panel, and then click "START NEW SUBMISSION" under Quick Links, or
- Click the [Submissions] icon on the left side panel, and then click "CREATE NEW SUBMISSION" located on the top right corner.

HOW TO CREATE A NEW SAMPLE SUBMISSION



- Submission Name: This is an optional field to help you identify your submission.
 Create a submission name that is complex enough to easily and uniquely identify each submission.
- 2. Type of Submission: Select "FFPE Tissue" from the drop-down menu. Only one sample type is allowed per submission.
- 3. Statement of Work: Select from the drop-down menu. Once selected, PI,
 Dissemination Recipient(s), and Scope of Work will be displayed. To make any
 changes to this information, please contact SOW@bcgsc.ca.
- 4. Sample Number: Enter the number of samples you are submitting. Unless otherwise instructed, samples must be submitted in 1.0 mL Matrix tubes (see sample "Submission Requirements" table on page 5) we do not accept other sizes of Matrix tubes.
- 5. If 1.0 mL Matrix tubes are available in your lab, click "No" under "Request a rack of 1.0 mL Matrix tubes", and then proceed to Step 7.







- 6. If 1.0 mL Matrix tubes are not available in your lab, pre-labeled tubes can be supplied by the GSC. For this request, follow these steps:
 - Click "Yes" under "Request a rack of 1.0 mL Matrix tubes".
 - You must use the "Prepopulate the form by uploading an Excel file" feature to submit Sample IDs and Tube Labels for your samples. Refer to Step 9 for more details.
 - Once Sample IDs and Tube Labels have been uploaded, we will receive your request and start preparing your tubes. Note that your online submission form will be locked while we are preparing your tubes.
 - When your tubes are ready to be picked up, we will send you the "Ready for Pickup" email notification. At this point, your online submission form will be unlocked, so you can proceed to complete the rest of the form.

Before unlocking your form, we will update your submission with Factory Barcode, Well Location, and Rac ID. **Do not change any entries in these fields.** If changes are necessary, contact GSC_Submissions@bcgsc.ca to see how you should proceed.

- 7. Taxonomy: You may enter multiple Taxonomy IDs per submission. Search for the taxa of your samples by typing NCBI Taxonomy ID (e.g., 9606) or name (e.g., human). Only selected Taxonomy IDs will be available later in the "SAMPLE INFORMATION" section under the Taxonomy field.
- 8. Sample Options: Select all that apply to your samples (Cell Lines and/or Xenograft). For these sample types, there will be additional fields to complete.

- 9. Prepopulate the form by uploading an Excel file: You have an option to prepopulate the sample information form by uploading an Excel file. If you answered "Yes" to "Request a rack of 1.0 mL Matrix tubes", the "Prepopulate the form by uploading an Excel file" checkbox will be automatically selected as you will need to use this feature to provide Sample IDs and Tube Labels.
 - Click the checkbox next to Prepopulate the form by uploading an Excel file:



- Click "DOWNLOAD TEMPLATE".
- Enter your sample information directly into the Excel file that is generated:
 - you can copy and paste entries from a pre-existing Excel file that contains your sample information;
 - » If a field has a drop-down menu, you must select from the available options;
 - » Some fields may require validation after the sample information form has been populated;
 - » All fields do not need to be completed in order to use this feature to prepopulate the sample information form; and
 - » Any missing or invalid fields can be corrected within the actual sample information form.
- Once completed, save the Excel file on your computer.
- Go back to the "Create a New Submission" section and click "UPLOAD EXCEL FILE".
- A dialogue window will open that allows you to navigate to the location on your computer where you saved the Excel file.
- The "SAMPLE INFORMATION" section will be auto-populated based on the entries in the Excel file.





- 10. Unused Sample: Indicate what you want us to do with any unused samples upon completion of your project. If you select "Destroy unused sample", we will discard your samples six months following data dissemination.
- 11. Click "START DRAFT" on the bottom right corner.
- 12. You can always go back to the "SET UP" section to edit the above fields:



SAMPLE INFORMATION FORM

Overview

- If you have requested a rack of 1.0 mL Matrix tubes from the GSC, your Sample
 Information Form will be locked until these tubes have been labeled and are ready to
 be picked up. Also note that we will update Factory Barcode, Well Location, and Rac ID
 before unlocking your form. Ensure that these fields remain unchanged.
- 2. Mandatory fields are denoted by *. Certain field selections will grey out mandatory fields. In these situations, field entry is not required. For example, if Tissue Disease Status is "Normal," Pathology will be greyed out, so you do not need to provide this information.
- 3. For additional information on any given field, click the icon next to the column heading:



4. There are tools on the top right corner to help you complete each field more conveniently:



- Paste Range: This option can be helpful when you have a pre-existing Excel file that you would like to copy and paste line items from. For the fields without dropdown menu, you can copy and paste directly into the form from your Excel file without using this tool.
- **Fill Down:** This option can be helpful when you have several consecutive rows that will contain the same value.
- **Clear Column:** This option can be helpful when you would like to reset values in the entire column.
- 5. Validator will give an orange warning message for any entries that we want you to double-check. If the information looks correct, you can proceed. You will receive a red error message when the entry is missing or incorrect. These entries must be corrected before you can submit your sample information form:



6. You can download the Excel file of your submission by clicking the "Download" icon next to the Submission ID:







Sample Information

1. Sample ID: This field is a unique identifier referring to the actual sample being submitted; you will receive your data back linked to Sample ID.

Sample ID Requirements

- Between five and 80 characters with no symbols and no spaces (hyphen & underscore are allowed);
- Sufficiently complex to help distinguish your samples from other samples; and
- If submitting two or more identical samples, indicate that they are identical by using the same Sample ID.
- 2. Tube Label: Enter "Tube Label" as it is written on the tube; can be the same as the Sample ID.

<u>Tube Label Requirements</u>

- · Less than 13 characters with no symbols (hyphen & underscore are allowed); and
- All labels must be unique to differentiate between tubes.
- 3. Taxonomy ID: Auto-filled from the "SET UP" section.
- 4. Factory Barcode: If you have a 2D barcode reader available in your lab, scan the 2D barcode on the bottom of your 1.0 mL Matrix tube for this information. If not, leave it blank. If your tubes have been pre-labeled by the GSC, we will fill in this field for you.
- 5. Well Location: Samples must be sorted by columns, so they are A1, B1, C1, etc. and NOT by row (i.e., A1, A2, A3, etc.). **If samples are not sorted by columns, additional rearraying costs may apply.** If your tubes have been pre-labeled by the GSC, we will fill in this field for you.
- 6. Tissue Fixation Process: If any fixative was used, you must fill in the "Tissue Fixation Process" field. For FFPE tissue, enter "FFPE." This is a type-ahead input field; begin typing and a list of suggestions will appear for selection. If necessary, you may enter free text.

- 7. Alternate Sample ID: This field is an optional identifier. The Alternate Sample ID must contain no symbols or spaces (hyphen & underscore are allowed) and be sufficiently complex to help distinguish your samples from other samples.
- 8. Patient ID: This is used as a common identifier in our database to link all samples received from the same patient; this assists with our QC processes as well as downstream analysis.

Patient ID Requirements

- A minimum of five characters with no symbols and no spaces (hyphen & underscore are allowed);
- Sufficiently complex to avoid unrelated individuals from having the same ID;
- De-identified; do not use hospital ID or PHN which may link back to the patient or patient records; and
- If two or more samples are from the same patient, they must have the same
 Anonymous Patient ID; do not add a prefix or suffix to differentiate normal/tumour
 pairs or different aliquots of the same sample.
- 9. Participant Study ID: This is an optional field.
- 10. Family Information: If applicable, enter any relevant family information such as a family history of disease or specific family relationship to other samples being submitted (e.g., proband, father, mother, sibling).
- 11. Developmental Stage: Enter the developmental stage of the sample being submitted (e.g., larvae, embryo, adult, pediatric).
- 12. Sex: Select from the drop-down menu. This information is helpful for QC processes and downstream analysis.
- 13. Collection Date: Enter the date that your sample was collected using the calendar (e.g., date of patient blood draw or date that a sample was collected in the field). The date must be in the YYYY-MM-DD format (e.g., 2022-01-24).





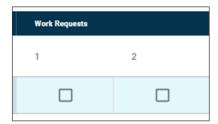
- 14. Anatomic Site: Enter the site from which the sample you are submitting was derived.

 This is a type-ahead input field; begin typing the anatomic site and a list of suggestions will appear for selection. If necessary, you may enter free text.
- 15. Tissue Disease Status: Select from the drop-down menu.
- 16. Disease Status of Patient (for Normal Samples): Enter the disease/health status of the patient. This information can help us track the disease status of the patient separately from the sample. For example, if you are submitting a normal skin sample from a patient with melanoma, the sample is normal but the disease/health status of the patient is "Melanoma".
- 17. Tissue Type: For diseased samples, select from the drop-down menu. Select "Benign" for benign tumour and "Non-neoplastic disease" if not cancer.
- 18. Cell Type: If a specific cell type has been used, specify the cell type. For example, if B-cells were sorted from lymph node, Anatomic Site would be "Lymph node" and Cell Type would be "B-cells." This is a type-ahead input field; begin typing the cell type and a list of suggestions will appear for selection. If necessary, you may enter free text. An orange warning message will display, but you can still proceed.
- 19. Collection Method: Enter the method used to collect the tissue. For FFPE tissue, enter "FFPE."
- 20. Scrolls or Cores: Select from the drop-down menu.
- 21. Number of Scrolls or Cores: Enter the number of scrolls/cores per tube. The recommended submissions amounts are:
 - Scrolls: 10 µm scrolls, minimum tissue surface area of 120 mm²
 For tissue surface areas ≥120 mm², one 10 µm scroll per Matrix tube
 For tissue surface areas ≤ 120 mm², up to 3 x 10 µm scrolls per Matrix tube
 For very small tissue, multiple tubes of 3 x 10 µm scrolls may be required
 - Cores: up to 2 cores in one Matrix tube, size 2.5 mm x (1-3 mm)

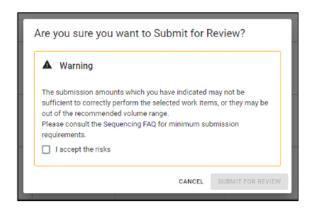
- 22. Scrolls Thickness (µm): Enter the scroll thickness in µm. Prior to preparing your samples, review the "<u>Frequently Asked Questions</u>" web page to ensure that your samples meet the requirements.
- 23. Pathology: This is a type-ahead input field; begin typing the disease name. A list of suggestions will appear for selection. The list is generated from <u>Disease Ontology</u>. If there are no appropriate matching selections, you may enter free text. **Do not use abbreviations.** An orange warning message will display, but you can still proceed.
- 24. Additional Pathology Info: For diseased samples, if applicable, enter pathology subtype/subgroup information such as molecular subgroup.
- 25. Pathology Occurrence: For tumour samples, select from the drop-down menu.
- 26. Pathology Grade: For tumour samples, if known, enter grade (e.g., G1, G2, G3, G4, Low, High).
- 27. Pathology Stage: For tumour samples, if known, enter stage by selecting from the drop-down menu.
- 28. Tumour Content (%): For tumour samples, enter percent tumour content of the sample, if known. It must be a specific number; please do not enter a range.
- 29. Treatment Status: Select treatment status at the time your sample was collected from the drop-down menu.

WORK REQUESTS & WORK REQUEST ASSIGNMENT

- 1. Please refer to your SOW to obtain the work request information that will be required in this section.
- 2. If the samples you are submitting have different library protocols and/or sequencing goals, you will need to click "ADD WORK REQUEST" on the top left corner to be able to enter different work requests for each sample.
- 3. Once the "WORK REQUESTS" section has been completed, you will need to assign one of the work requests to each sample in the "WORK REQUEST ASSIGNMENT" section.
- 4. Assign a work request to each sample by clicking the checkbox under the appropriate "Work Requests" column:



- You can edit work request details by going back to the "WORK REQUESTS" section.
- If the same work request should be applied to all the samples, simply click the first checkbox, then click "FILL DOWN" on the top right corner.
- 5. The required amount of tissue is validated based on the library protocol selected. If you are submitting less than the minimum amounts, you will be asked to accept the risks of failure before you can submit your sample information form for review:





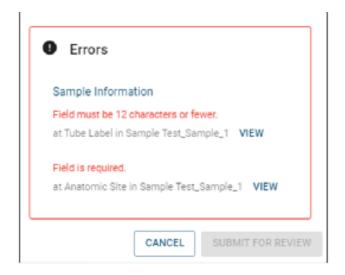


COMMENTS & PLATE INFORMATION

- 1. Use the "COMMENTS" section to:
 - Enter any additional comments regarding your work request, such as specific pooling strategy, should any clarifying information be required; and
 - Provide any extra information or details regarding your submission or samples.
- 2. For your tube rack for FFPE tissues, please provide Plate ID and Rac ID (if applicable) in the "PLATE INFORMATION" section:
 - Plate ID is the name your lab assigns to the tube rack. You may handwrite your
 SOW number, project name, etc. on the rack and use that as Plate ID;
 - If your samples are being submitted in pre-labeled Matrix tubes provided by the GSC, Plate ID may not be identical to Rac ID, as Rac ID is the internal name used by the GSC; and
 - If you have received a rack of 1.0 mL Matrix tubes from the GSC, Rac ID will be filled in for you. You can also find this information in the label affixed to the rack.

SUBMITTING THE SAMPLE INFORMATION FORM

- 1. Once you have reviewed all the fields and are satisfied with the sample information form, click "SUBMIT FOR REVIEW" on the top right corner.
- 2. If any of the samples do not meet the minimum recommended amount, click on the "I accept the risks" checkbox in order to proceed.
- 3. If there are any errors that need to be addressed, you will see the following message:



Click "VIEW" and you will be taken directly to the cell whose error must be fixed before submitting.

4. Once your sample information form has been successfully submitted, you will receive "Sample Information Form Received for Review" email notification.





AFTER SUBMITTING SAMPLE INFORMATION FORM FOR REVIEW

1. Your sample information form will be reviewed for completeness and accuracy. You will then receive "Edits Requested" or "Submission Approved" email notification.

For "Edits Requested":

- In order to resubmit the form, you must confirm or amend all of the fields which
 have been highlighted yellow or orange by the reviewer. As you address each field
 in the form, its colour will change to green or blue. When there are no more yellow
 or orange fields left, you can resubmit;
- Some fields may also contain a comment made by the reviewer, in which case those fields will be marked with a small red triangle. You can hover your mouse over those fields to see the reviewer comments; and
- You also have the option of making edits to any field, including those which have not been highlighted by the reviewer. Any such fields will become highlighted purple, and are subject to final review by the GSC Submission team.

Below is a legend of the highlighting colours you may encounter while making edits:

- » Yellow: Needs Attention—the reviewer believes an edit may be required.
- » Orange: Changed by Reviewer—the reviewer has already changed the value of this field, and would like you to confirm (or re-edit). Hover over the field with your mouse to see the change that has been made.
- » **Blue:** Amended by Submitter—to be reviewed by the GSC Submission team.
- » Green: Confirmed by Submitter—subject to final review by the GSC Submission team.
- » Purple: Additional Edit by Submitter—subject to final review by the GSC Submission team.

You should use the following three modes ("Edit", "Highlight", and "Read-only") when addressing reviewer comments and changes:



- Edit: This is the default mode. When in 'Edit' mode, you are able to alter any field to change its value. Whenever you have made an edit to a field, it will turn blue or purple.
- » **Highlight:** When you switch to 'Highlight' mode, clicking on a field should turn it green. You may highlight a field green to indicate that you agree with an edit made by the reviewer, or that you believe the highlighted field already has the correct value (clicking on a field multiple times while in highlight mode will toggle between 'unconfirmed' and 'confirmed' state).
- » **Read-only:** All error and warning messages will be hidden.
- 2. Once your submission has been approved, you will receive a "Submission Approved" email notification with detailed instructions for dropping off or shipping your samples.







SAMPLE SUBMISSION BY COURIER

Once the sample submission form is approved, samples should be shipped on dry ice and addressed to:

Dr. Andrew Mungall - Biospecimen Core, Room 508
Genome Sciences Centre BC Cancer
Suite 100 - 570 West 7th Avenue
Vancouver, BC Canada
V5Z 4S6 (*If shipping via Purolator, use V5Z 1B3)

email: amungall@bcgsc.ca

Tel: 604-707-5900 ext 675411

When samples have been shipped, we ask that you please email sampleshipments@bcgsc.ca to notify us of your shipment and the associated tracking number so we can monitor the progress during transit.

If shipping on dry ice, please ensure that there is sufficient dry ice for a couple of days. We recommend shipping Monday to Wednesday as we cannot accept packages on weekends.



SAMPLE DROP-OFF

The sample submission form must be reviewed and approved by GSC personnel prior to submitting samples to the GSC.

Regular hours for sample drop-off and plate pick-up:

Monday - Friday: 9:30-11:30 am and 1:30-3:30 pm

Location:

Suite 100-570 West 7th Avenue, Vancouver, BC V5Z 4S6

To enter the building, dial #100 on the intercom and the receptionist will let you in. The reception is on the ground floor (past the elevators and on the left). Go through to reception and ask the receptionist to call or page anyone from the Biospecimen Core group. We'll come down to reception to meet you.

Technology Platform

Echelon Innovation Centre 100-570 West 7th Avenue Vancouver, B.C. V5Z 4S6

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Research Department

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