

gDNA submission guidelines for whole genome (WGS) or exome sequencing

How to submit your gDNA samples?

- Fill in the gDNA **submission excel file** found on our website and email it to us. This must contain all relevant information, no information can be transmitted only within the email text or orally.
- In parallel to submitting the form, please bring DNAs on ice during regular office hours. If we haven't received your samples yet when starting processing a project, the project will be delayed. DNAs should be on ice, in **DNase-free PCR-grade 0.5ml-2ml microtubes**, but not in PCR tubes/strips/PCR plates. Leave some space on the side of tubes for GECF codes.
- Indicated tube **labels** must match exactly the ones on your physical tubes. If available, labels of profiles must also match perfectly either tube labels or sample names.
- Avoid using "S1", "S2"... in your samples name. These codes are already appended by the sequencer to fastq files, which causes confusion.
- If you need to **modify** or comment your submission in any way, do it by email, even when you have already told us orally.

Recommendations for gDNA preparation

- If planning for a variants calling experiment, include a **control sample** that will serve as a reference.
- Avoid **bacterial contamination**, for instance during dissection or FACS sorting (when using old contaminated buffers).
- Perform the **RNase treatment** that is often only optional in mainstream DNA extraction kits. RNAs can heavily contaminate gDNA prep and may affect gDNA quantification and other downstream procedures.
- Nanodrop measurements of **gDNA concentration** may be very inaccurate, in particular at low concentrations (<20 ng/ul). Therefore, always report 260/280 and 260/230 ratios when nanodrop is used. Alternatively, a more accurate and sensitive method is Qubit DNA HS.
- Depending on your project/aims, the lower limit to the **gDNA quantity** required for library prep differs. Thus contact us in advance to discuss it.
- If DNA samples come from **FFPE** (paraffin-embedded samples), contact us in advance to discuss the DNA extraction protocol.
- **DNA elution** buffer of choice depends on your extraction method and downstream application. Efficient elution in general requires a basic pH. In addition, some EDTA is often present to increase stability of DNA during long term storage, but too much EDTA (1mM as in regular TE) can inhibit certain downstream applications. Here are some considerations when choosing elution buffers or H2O (all these reagents/H2O should be PCR-grade):

- "low-EDTA TE" (10mM Tris, 0.1mM EDTA) at pH8, is probably the safest choice as it allows efficient
 elution in most case (check your extraction documentation), while incorporating some EDTA to
 favor long term DNA storage, but in a sufficiently low concentration to avoid issues with any
 downstream application.
- Qiagen AE buffer (10mM Tris, 0.5mM EDTA, pH 9) is fine for most downstream applications, but ask us in advance in case of doubts.
- 10mM Tris pH8 is fine as well.
- H2O is excellent for downstream applications, but check its pH as commercial/distilled H2O is in general acidic, hence not suitable for efficient elution with most extraction kits.
- **DNA storage** should be done at -20°.

Determining library prep method and sequencing parameters

- The choice of a library prep method depends directly on results from DNA quantification and QC, and is a shared decision between the user and the GECF. This discussion will address these topics:
 - Are you interested in sequencing whole genome or only exome? Exome-seq reduces a lot sequencing costs. If exome-seq is enough, do you also need UTR or promoters or custom regions such as a transgene?
 - If your organism genome size is small and thus a small sequencing output is sufficient, the **MiSeq** may be used. In that case, we will use the very long reads option available on that instrument (2x300nt), and library size will have to be adapted during library prep.
 - Are your gDNAs likely to contain bacterial DNAs (gut samples, neutrophils fighting an infection...)?
 - What will be the **lowest SNP/mutation frequency** that you will want to call? This will determine required coverage, hence read length and mio reads/sample.

Disclaimers

- The GECF guarantees successful library preparation when using a commercial method starting with gDNAs of concentrations and quality matching that method's specifications. In case a library prep fails or does not lead to satisfactory results due to technical problems, the GECF will prepare it again without additional charges. Yet, when it turns out that the issue does not come from the GECF (e.g. problem is reproducible, and/or a control library is successfully prepared in parallel), the libraries prep will be billed to user. Typical, but very rare, examples of GECF-independent issues are:
 - Library prep is inhibited by a contaminant present in the gDNA samples. These can be difficult to detect and can potentially harm library preparation.
 - A wrong gDNA concentration was indicated by user. Of note, in case of suspicion of a
 quantification issue and when sample quantities allow, we independently check user-provided
 concentrations. Yet sometimes sample quantities do not allow for this double-checking.
- When the GECF and users agree to perform an exploratory method, or to process samples even though they do not comply with recommended specs for the chosen method, the GECF cannot be held responsible for failures or unsatisfactory results (outside obvious errors from the GECF side).

- The GECF is an open access facility, therefore we cannot give guarantee regarding the confidentiality of data. If this is an issue, the easiest solution is to give codes to your samples. Upon request we can create access-controlled folders for data delivery.

Versions log

- v1.01: initial release.
- v1.02: clarified the advantages of the MiSeq
- v1.03: Modified significantly the accompanying excel sheet. Clarified that when submitting several independent projects, different submission forms should be used. In case these projects should still be sequenced together, it should be indicated as a comment. Also indicated that tube labels must match exactly the ones on your physical tubes. If available, labels of profiles must also match perfectly either tube labels or sample names. Clarified that when one needs to modify or comment a submission in any way, it must be done by email, even when the information has already been given orally.
- v1.04: Clarified to avoid using "S1", "S2"... in your samples name. These codes are indeed already appended by the sequencer to fastq files, which could cause confusion.
- v1.05 (03.11.2025): minor changes. Mentioned to specify in case a transgene needs to be sequenced for exome-seq. Clarified that the submission form itself must contain all information. Clarified that DNAs must be brought to GECF in parallel to form submission. Mentioned that people can request a lab-specific access-controlled data delivery folder.