

Genomic DNA, Targeted Sequencing and Bisulfite Sequencing Submission Guidelines

DNA Extraction

The Genomics Core does not recommend one specific procedure for isolating DNA, but rather suggests that if the lab is already working successfully with DNA in other molecular biological applications that they continue to use the same method. If purity and/or quality issues should arise, Genomics Core staff can provide alternative recommendations.

DNA Quantities per Assay

Genomic DNA : 100 ng–2 μg of high molecular weight DNA **Targeted captures—exome, methyl, custom**: 3 μg of high molecular weight DNA preferred and recommended; 200 ng acceptable for limiting samples **Whole Genome BS and RRBS**: 4-5 μg of high molecular weight DNA

Please provide 3-4 μ L extra for QC purposes. The concentration of your DNA ideally should be 20-100ng/ μ L, provided in no less than 20 μ L, and a maximum volume of 50 μ L.

DNA samples should meet the following requirements:

- 1. It is **strongly** recommended that all samples be treated with RNase prior to submission to ensure samples are free of RNA contamination. RNase treated samples must also be cleaned up post treatment.
- The A₂₆₀/A₂₈₀ should be 1.7-1.9. A ratio <1.8 is often indicative of protein contamination. A ratio >1.8 indicates RNA contamination or residual phenol or beta-mercaptoethanol. Protein contamination should be removed by re-extraction using phenol:chloroform:isoamyl alcohol or another pass over a commercially available column. Other contaminants can be cleaned up by EtOH precipitation.
- 3. Samples will preferably be quantified using PicoGreen and either a plate reader or Qubit fluorometer. If these are not available, samples may be quantified on a spectrophotometer or NanoDrop. Please provide extra sample in this case, as spectrophotometers are known to overestimate quantity, often severely.
- 4. Samples should be in 10 mM Tris-HCl or nuclease-free water. If sample(s) is not in water, note the type of buffer on the submission form.
- 5. 100 ng aliquot of each sample should be run on a 1% agarose gel to check integrity, and a labeled gel picture should be provided when samples are submitted. Intact samples should have a single band above the highest marker band, with no/minimal smearing evident.

Shipping/Sample Drop-off

- 1. An iLab request should be filled out and approved prior to submission.
- 2. Samples <12 should be submitted in either a 0.5 mL or 1.5 mL nuclease-free microfuge tube with the sample name **clearly** written on the cap—no more than 6-8 letters/numbers—**and the PI's name on the side**. If >12 samples submitted, the samples should be in a nuclease-free PCR plate sealed appropriately with the PI name on the side of the plate skirt (spreadsheet with plate layout should be e-mailed to <u>genomics@vai.org</u>and should also accompany plate). All items shipped should be sealed in a plastic bag.

Sample should be shipped on dry ice overnight to

Van Andel Institute Attention: Marie Adams 333 Bostwick Avenue, room 2011 Grand Rapids, MI 49503