BS-Seq sample preparation guidelines

Genomic DNA for BS-Seq and RRBS-Seq

IGATech offers nucleic acids extraction service and we can set up a dedicated extraction workflow for your specific substrate. Please enquire.

Prepare the DNA following your favorite extraction method, even if we strongly recommend to use commercial column-based protocols. Control your DNA on 0.8% agarose gel to check for integrity.

Submit minimum of 1 µg of DNA per sample (minimum concentration of 20 ng/µL). Please note that fluorimetry-based quantification (*e.g.* Qubit, plate-reader) assays are more accurate methods than absorbance-based methods (*e.g.* Nanodrop) which might overestimate the quantity.
Quality of the DNA should be 260/280 > 1.8 and 260/230 >1.8.

DNA has to be resuspended in 10mM Tris-HCl pH 8.5 (standard elution buffer of most commercial column-based extraction kits); water is accepted as an alternative (**NO high concetration of EDTA must be present in the solution** – e.g. TE buffer has 1mM EDTA - but consider 10mM Tris-HCl as best buffer for HMW DNA stability or 10mM Tris-HCl + 0.1 mM EDTA.

The tubes must have, on the vial top, a clear and permanent sign (or a thin label) with a progressive number of the mailed samples and the customer's name (at least the initials).

General best practice and quality for DNA:

- *260/280>1.8 and 260/230>1.8*
- *Quantification made by dsDNA-specific fluorimetry (Qubit/fluorimeter)*
- *Avoid repeated freeze-thaw cycles (use aliquots)*
- *Do not expose to high temperatures (>65 C)**
- *Store the DNA in stabilizing buffer (Tris-HCl pH 8.0-8.5)*
- *Do not provide DNA with EDTA (conc. > 0.1 nM)*
- *DNA must be RNA-free (so RNAse treatment is strongly suggested)*

- *Does not contain phenol, polyphenols*

**Especially for HMW DNA*

Mail DNA samples in 1.5 or 2 mL Eppendorf tubes sealed with parafilm (0.5 mL / 0.2 mL tubes will not be accepted).
If you have 24 or more samples, please put them in a 96-well skirted plate sealed with adhesive/heat-sealed aluminum foil. The tubes must have, on the vial top, a clear and permanent sign (or a thin label) with a progressive number of the mailed samples and the customer's name (at least the initials).
Send DNA samples in a cold pack (*e.g.* Blue ice) or dry ice. Do not ship plates without secondary containment as these may crack when placed directly on dry ice.

Please, do not forget to send us compiled **Samples Spreadsheet** and the **Customer registration form**, together with shipped samples as well as via e-mail.