



Exome-Seq sample preparation guidelines

Genomic DNA for target/exome sequencing

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

The quality of the DNA sample can have a significant effect on the successful of the experiment both using SureSelect (Agilent), MedExome (NimbleGen) and Nextera (Illumina) enrichment systems. Poor quality DNA can determine the presence of duplicated (e.g. clonal) reads and consequent insufficient coverage.

Recommended commercially available kits

DNA extraction kits (catalog number)	Blood*	Tissue (incl. tumors)	Cultured cells
Qiagen DNAesy Blood & Tissue Kit (69504)	X	X	
Qiagen Genra Puregene Cell Kit (158722)			X
Qiagen Genra Puregene Blood Kit (158445)	X		
Qiagen QIAmp DNA Blood Midi Kit (51183)	X		

* Use preferentially EDTA or ACD as anticoagulant. Heparin can inhibit Taq Polymerase.

Submit minimum 200 ng of DNA for Illumina and 1 µg for Agilent and Roche enrichment protocols. 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 must be resuspended in 10 mM Tris-HCl pH 8.5 (standard elution buffer of most commercial column-based extraction kits); water is accepted as an alternative (no EDTA must be present in the solution – e.g. TE buffer).

A minimum concentration of 100 ng/ μ L with majority of the fragments >20Kbp (perform a 0.8% agarose gel check to control for possible degradation).

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. 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).

Please, do not forget to send us the compiled **Samples Spreadsheet**, both with the shipped parcel and via e-mail. In order to be able to properly track and safeguard your samples we also ask you to send us the **Tracking Number** via e-mail.