City of Hope

Sample Requirements

[Sample Submission Guidelines](http://www.cityofhope.org/integrative-genomics-core-sample-submission-guidelines)

You are encouraged to set up a meeting to consult for experimental design with the core director, Dr. Xiwei Wu, to determine the optimal platform, library preparation protocol, and sample size. Please fill out the service request form at the time of sample submission. Typical turnaround time for microarray service is one - two weeks, and three - four weeks for sequencing, one - four weeks for data analysis. Please note that actual turnaround time might vary depending on the work load and complexity of the project.

**Sanger Sequencing Sample Requirements:**
Log on the LIMS system at [dnatools.coh.org](http://www.cityofhope.org/submit-a-dnatools-order)

**Concentrations:**

* Template - PCR Product:

Concentration at least 0.5 ng per 100 base pairs for fragments less than 500 bp.

* Dilute PCR sample to 1 ng/ul if PCR fragment is less than 500 bp.

If your PCR sample is greater than 500bp then dilute PCR to 3ng/ 100 base pairs in 2 ul volume.

For example: If your PCR size is 600 base pairs, multiply 600 X 0.03 which will give you the total amount of DNA necessary ( 18ng) for sequencing. Then dilute your PCR sample to the correct working concentration of  9ng/ul in 2ul total volume.

* Template – BAC Clones

1ug/ ul concentration for BAC DNA

Template – Plasmid

* Concentration at 100 ng/ul

2.0 ul needed per reaction

* Custom Primers

Concentration at 1pmol/ul
10 pmol/ul for BAC sequencing primers
4ul needed per reaction

**Entry Form** (After you have clicked Enter DNA Sequencing Requests, and entered the number of reactions)

* For City of Hope users, leave the PO Number field blank.  This is for outside users only.
* Under the Grant Code Num. field please enter your grant code.
* The account number you enter must use the subaccount of 8028. Do not change this.  Enter your entire account number (20/30 – 8028 – 6 digit account number) Ex. 30-8028-xxxxxx
* Enter the PI name “last name, first name” and in all capitals.  Ex: SMITH, JOHN

**Chemistry**

* BigDye\_V3.1 – For basic sequencing.  Can read out to approximately 900 base pairs
* BigDye\_V1.1 – Used only for short (less than 500 base pairs) PCR products when high resolution near the priming site is needed
* • BigDye\_3.0-dGTP – Used only for templates that are likely to form strong secondary structures, hairpin loops, or with high GC content (greater than 75%) or bisulfite-treated samples.  When using this chemistry, please include the Tm of your primer in the comments box

Make sure after submitting the entry form, you print 2 copies of the table that appears. One of these copies must be placed in the inbox to inform us that the samples are ready to go.  If we do not receive a copy of this form, your samples will never be sequenced.  The other copy is for your reference.

Please label your samples with less than 8 characters if possible.  Write the labels on the cap of the tubes you are submitting. **Make sure that the labels on your tubes matches identically to the labels on the order form.**

If you need a Chromatograph viewer, you can download one from [Life Technologies.](http://www.lifetechnologies.com/order/catalog/product/4474779)