



# DHVI Viral Genetic Analysis Core

## Mission

The Duke Human Vaccine Institute Viral Genetic Analysis Core facility provides sequencing (NGS and Sanger) and genomic technologies to serve the needs of the Duke Human Vaccine Institute, Duke Center for AIDS Research, Duke University and external collaborators.

## Leadership / Experience

- **Director:** Todd Bradley, PhD
  - Dr. Bradley is a Medical Instructor in the Department of Medicine and became facility director in 2016. Dr. Bradley has over 10 years of experience in developing next-generation sequencing strategies to investigate mechanisms of cell biology and immune cell responses
- **Manager:** Bhavna Hora, PhD
  - Dr. Hora is a researcher and a core manager with over 17 Years of experience in molecular biology, immunology and virology. Dr. Hora has been managing the sequencing core for the past two years and has over 12 years of experience in HIV sequencing, SGA, Q-PCR, mutagenesis etc.

## Services

- **Next-Gen Sequencing:** High Throughput RNA/ DNA sequencing, library prep, NGS runs and data analysis
- **10X Genomics:** The Chromium Single Cell 3' Solution provides high-throughput, single cell expression measurements that enable discovery of gene expression dynamics and molecular profiling of individual cell types.
- **Sample and library QC:** Quantification and quality control of samples and libraries
- **Single Genome Amplification:** Isolation and analysis of sequences for single nucleotide polymorphisms within the HIV-1 genome
- **Sanger Sequencing:** sequence up to twenty-four 96-well plates with reads of up to 1000 base pairs per sample
- **PCR Purification:** utilizing an innovative paramagnetic beads technology for high-throughput purifications of PCR amplicons
- **Site Directed Mutagenesis:** single, multiple base, deletion mutants in a plasmid
- **Drug Resistance Mutations:** Population sequencing of viral samples to determine the DRMs in the sample
- **Sequence Analysis of Viral Stocks:**
- **Co-Receptor usage Assay:** Determine coreceptor usage of a virus sample in NP2 cells by measuring p24

## Instrumentation and Technology

### Next-generation (NGS) Sequencing on Illumina



MiSeq



NextSeq

We offer services on Illumina MiSeq and NextSeq 500. Because of their similar chemistry, same library preparations can be used across these instruments, allowing us to tailor to researchers' sequencing needs according to turn around time (few hours to three-day run) read length (75 bp single-end reads to 300 bp paired-end reads) and throughput (300 Mb/run to 500 Gb/run).

### Single Cell Genomics



The Chromium Single Cell 3' Solution provides high-throughput, single cell expression measurements that enable discovery of gene expression dynamics and molecular profiling of individual cell types.

### Sample/ Library Quantification and Analysis



StepOne Plus (qPCR)



Qubit



Tapestation 2000

### Sanger Sequencing and PCR Purification



ABI 3730XL DNA



Biomek FXP Laboratory Automation workstation

## Recent Projects / Publications

- Used **high-throughput immunoglobulin sequencing (Ig-Seq)** to determine the dynamics of neutralizing antibody response to HIV infection (Human; Bonsignori et al. *Sci Transl Med.* 2017) and HIV vaccination (rhesus macaques; Zhang et al. *Sci Transl Med.* 2016; Humanized mice (Verkoczy, Haynes *in prep.*).
- Performed **site directed mutagenesis** of antibody and HIV envelope genes to determine important functional mutations (Bonsignori et al. *Cell.* 2017; Wiehe et al. *submitted*, 2017.).
- **Sequencing HIV-1 genome** from global sites (Liu et al. *Retrovirology.* 2017; Chen et al. *Plos One* 2016.).
- **Single cell RNA-seq** of blood cells from neonatal and adult macaques to determine transcriptional differences before and after vaccination (Han et al. *in prep.* 2017).

## Reservations / Service Requests

The DHVI VGA Core offers 3 ways to utilize our facility to accomplish your research objectives:

- **Sample submission-** Researchers provide DNA/RNA or PCR amplicons and service is performed by Core staff (Sanger DNA sequencing, PCR amplicon purification and Illumina next-generation DNA/RNA sequencing, HIV co-receptor, subtype and drug resistance analysis).
- **Intermediate use-** For Illumina next-generation DNA/RNA sequencing submit prepared libraries and Core staff can QC, optimize and provide appropriate sequencing kits and generate raw data
- **Independent use-** Researchers can reserve and operate the instruments independently and provide all of the required reagents for Illumina Miseq, Illumina NextSeq, real-time qPCR, TapeStation, Qubit fluorometer.
  - Complete training in the independent operation of the Illumina and QC platforms is required

### First step:

- Contact Core personnel for consultation and a price quote for your desired service. We will guide you through the process to achieve your research goal as fast and cost-effective as possible.

### Second step:

- Link user NetID's to the appropriate PI and fund codes in **CoreResearch@Duke** system for integrated reservation and billing services.
- Make Reservations OR Submit requests for any Core service

## Contact Us

**Website:** <https://shared-resources.dhvi.duke.edu/dhvi-core-facilities/dhvi-sequencing>

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