FACILITIES AND OTHER RESOURCES
University of Idaho

IBEST Genomic Resources Core Facility
Recent advances in DNA sequencing and transcriptome analysis have ushered in a new era of biological inquiry. To take advantage of these technologies IBEST has developed a state of the art core facility so researchers can study biological processes at the genome-scale. To enable the use of these technologies the IBEST Genomics Resources Core not only provides access to technology, but access to staff with the experience and expertise in molecular biology methods and bioinformatics needed to acquire, analyze and visualize data generated from high throughput technologies in genomic research.

Core Facility Infrastructure
The Genomics Resources Core Facility has equipment necessary to generate data for applications including DNA and RNA sequencing, high throughput sample preparation and quality assurance, and computational resources described below. In addition, there are thermocyclers, centrifuges, microcentrifuges, freezers, refrigerators, gel-imaging systems, a vacuum-centrifuge, and computational resources. The Core facility occupies approximately 780 sq. feet of laboratory space in the newly finished Integrated Research and Innovation Center (IRIC) room 210, and 620 sq. feet of laboratory space in IRIC 142, with approximately 300 sq. feet of office space in IRIC rooms 224 and 226 at the University of Idaho main campus in Moscow, Idaho. The Core facility infrastructure is described in more detail below.

DNA and RNA Sequencing
Sequencing has become an indispensable tool for basic biological research, biomedical research, diagnostics and biological systematics. Current applications using sequencing include whole genome shotgun sequencing (including *de novo* sequencing of previously unknown genomes), transcriptome and mRNA sequencing, targeted re-sequencing, single nucleotide polymorphism (SNP) discovery, amplicon sequencing for studies on microbial community composition and phylogenetics, and many other applications. Equipment for sequence generation includes:

Pacific Biosciences Sequel II
The Sequel II represents the latest in 3rd-generation long read sequencing technology. When samples prepared for HiFi sequencing, yields of up to 4 million high quality (>99% accuracy) reads 9-13Kb in length are possible. Libraries prepared for Continuous Long Read sequencing can produce reads up to 170Kb in length, and yields approaching 160Gb. Data produced by this sequencer can be used for a variety of applications, including *de novo* genome assembly, genome resequencing to call variant (SNVs, InDels, SVs, and CNVs), RNA Sequencing (Iso-Seq) for genome annotation, and for long-amplicon sequencing. PacBio sequencing technology has no GC content bias, and does not require amplification of DNA resulting in uniform coverage. Additionally, PacBio sequencing technology enables detection of methylation during sequencing, allowing researchers to study both the DNA sequence and the pattern of methylation simultaneously. Sequencing costs for the Sequel II are much lower per Gbp than for previous long-read sequencing technologies (e.g. Sequel I, RS II), enabling new types of research at larger scales and making long-read technology affordable for non-model organisms and projects with modest

budgets.

Illumina MiSeq
The MiSeq is the only fully integrated personal sequencer, delivering a rapid sample prep protocol and a full sequencing run in less than 56 hours. The MiSeq offers capabilities of up to 2x300bp paired-end sequences (600bp total per read) and 20Gb per run. Experiments that warrant the extra capacity from an Illumina HiSeq can also be conducted within the GRC core facility, where Illumina libraries are prepared in the core facility and sent to another core facility for sequencing. The GRC handles all of the communication with the external facility, and at the end of the process data are sent back to the GRC and the researcher.

Fluidigm Access Array and Juno Systems
The Juno and Access Array Systems provide PCR based, high-throughput, target-enrichment designed to work with all of the major next-generation sequencing instruments. These systems enable the user to enrich multiple unique targets (such as exons) from a large number of samples,simultaneously by using micro-fluidics and PCR. This results in quality results while minimizing the time, cost, and labor required for targeted re-sequencing projects. The Access array is capable of amplifying up to 480 loci in 48 samples simultaneously, while the Juno can amplify as many as 2400 loci in 192 samples simultaneously.

Apollo 324 for NGS Library Preparation
The Apollo 324 is a benchtop system that automates next-generation sequence library preparation workflows (Illumina TruSeq/Nextera libraries) by using bead technology to execute high-performance isolation and purification.

Advanced Analytical Fragment Analyzer
The Fragment Analyzer is a 12-lane electrophoresis array that provides accurate fragment quantification and qualification. It can quantify genomic DNA, total or purified mRNA, and prepared libraries in a range of sensitivities.

High Throughput Sample Preparation and Quality Assurance
Access to equipment for high throughput sample preparation gives investigators abilities for increased throughput and reduction of sample-to-sample variability over manual methods. In addition to providing high-end equipment for generating sample data, the Core enables researchers to perform accurate assessment of sample quality before committing extensive financial resources to a project. Equipment for sample preparation and QA in the Core include:

Kingfisher Flex: The Kingfisher Flex is a bead-based sample preparation system that eliminates pipette steps and the resulting waste from its protocols. It is a fully automated, scalable, and customizable system that delivers rapid and consistent results for RNA and DNA.

*Qiagen QIAxcel***:** The QIAxcel system is a multicapillary electrophoresis system designed to overcome the bottlenecks of gel electrophoresis. The fully automated system can process up to 96 samples per run.

*Molecular Devices SpectraMax Paradigm and Gemini XPS***:** The SpectraMax

Paradigm is a multimode microplate detection platform. It is the only user upgradeable microplate reader on the market that allows for real-time system configuration. Current cartridges include the absorbance (ABS) detection and the tunable wavelength (TUNE) detection cartridges. The Gemini performs nucleic-acid quantification, microbial growth assays, ELISAs, and reporter-gene assays to name a few.

Fluorometry: The DNA or RNA in samples can be quantified using fluorescent assays. For multiple samples in a 96-well plate format these assays can be done using the Gemini XPS microplate reader from Molecular Devices, smaller numbers of samples can be assayed using the Qubit 2.0 from Life Technologies.

Covaris Ultrasonicator 2200: The ultrasonicator rapidly shears DNA to a specified size while maintaining a consistent size distribution for any number of samples. It is invaluable for protocols requiring precise and replicable DNA fragment sizes.

Aurora System: A revolutionary platform for nucleic acid extraction based on a powerful electrophoretic purification technology from Boreal Genomics. The electrophoretic extraction technology is proven to purify DNA and RNA from extremely low abundance and heavily inhibited samples including soils from the Atacama Desert, Antarctic tundra, sea sediments, oil sands, and stool.

StepOne Plus qPCR instrument: Accurate quantification of sequenceable library is essential for a successful sequence run. This instrument allows for rapid and accurate quantification of Illumina libraries, thus improving cluster management on the flow cell and sequence quality.

Sage Science BluePippin: The BluePippin allows selection of high molecular weight DNA for long-range genomic applications such as genome sequencing.

Coastal Genomics LightBench MK II: Fast, easy, affordable, accurate, and precise automated agarose-based size-selection of DNA/library from 100bp to >20,000bp using real-time analysis and dual-colored fluorophores .

Bioinformatics Analysis Resources

The GRC offers bioinformatics support through staff bioinformaticians and can perform a full range of analysis tasks to address biological questions in areas such as population genetics, genomics, microbial community dynamics, functional genomics and systems biology. GRC bioinformaticians begin with raw data output from the instruments and proceed to quality assurance, data processing and analysis. Results are shared with the collaborator through a dialog that includes data interpretation and visualization. Analyses are done using software tools available in the public domain or developed by the GRC staff when project specific methods are needed. Core personnel have developed analysis techniques and pipelines for targeted assembly, transcriptome assembly, population variant analysis, SNP/INDEL detection, and RNA-seq analysis. These pipelines transform raw data into a form and format that can be mined by investigators. Throughout the data analysis process, a dialog is maintained with the investigator ensuring that the project meets its goals, figures are generated and summary tables are provided in a form that answers the biological question is useful for publication. As needed, the Core staff provides investigators with detailed knowledge of the laboratory protocols and bioinformatics methods used so they can be included in

reports and publications. The close collaboration between core staff and investigators often warrants inclusion of core staff as co-authors on publications.