



Services for R&D available at CAAD

Last update July 29, 2020

CAAD is a Research Infrastructure that provides resources and services to foster the innovation potential of Scientists, public users and Enterprises.

To reserve the following instrumentations/services, it is necessary to be registered with the "Agendo" centralized management system. To register, fill the Registration Form at the following link <u>http://caad.uniupo.it/servizi</u>.

If you are already registered, log into the system https://upo.agendo.science/.



Advanced Microscopy

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Overview

The Advanced Microscopy (AM) Unit provides expertise and tools for the analysis of the structural and molecular organization of both live/fixed cells and tissues, and time-lapse investigations. The service allows access to state-of-the-art optical microscopy instruments including transmitted light, fluorescence and confocal instruments, providing training and support for the correct use of the tools.



This service is intended for both members of the University of Piemonte Orientale and external user and companies.

Services:

This Unit offers:

- Support in experimental design
- Training of new users
- Live imaging/time-lapse acquisition and analysis
- Multi-color image acquisition and 3D rendering on images acquired by the confocal microscope
- Support for image processing and quantitative analysis of data

Equipments:

- Leica THUNDER Imager Live Cell
 - Fully motorized inverted fluorescence microscope DMi8
 - o Autofocus
 - o Objectives: 5x/20x/63x(oil)
 - o Filter Cubes: DAPI/FITC/TxR/Cy5
 - Camera: Leica DFC9000 GTC
 - Cage incubator
- Leica TCS SP8 confocal laser scanning microscope
 - Fully motorized inverted fluorescence microscope DMi8 CS Bino
 - o Autofocus
 - o Objectives: 10x/20x/40x(oil)/63x(oil)
 - o Filter Cubes: DAPI LP/FITC LP/RHOD LP
 - o DIC system accessories for 40x & 63x objectives
 - Stage incubator
 - Laser Lines: 405/458/476/488/496/514/561/633 nm
 - 4 detectors:
 - o 2 PMT



o 2 HyD GaAsP



Bioinformatics

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Overview

The Bioinformatics (BINF) Unit @CAAD is aimed at supporting and analyzing data in the genomic field, both for UPO researchers as well as for external laboratories and partners.

The advent of inexpensive high-throughput technologies allows the production of large-scale genomic dataset in practically all life-science domains.

Once produced, the data must be analyzed, very often in highly specialized manners, to extract useful information.

This requires a large set of advanced computational and statistical skills and the ability to communicate constructively with different interlocutors, in order to meet the specific questions of each research group.

BINF supports research in designing, conducting and interpreting high-throughput experiments in several life science domains.

We can operate in terms of scientific collaboration and/or in terms of fee-based services.



Services:

BINF Unit offers computational support for all the main type of high-throughput NGS sequencing analysis, including analysis of ChIP-seq data, mRNA and microRNA-seq data, exome sequencing data.

The computational support offered is mainly based on a local implementation of state-of-the-art NGS analysis pipelines. In addition to that, the group offers consultancy for non-standard analysis and support over long-term projects.

Infrastructures:

The Unit is equipped with last-generation desktop Linux computers as well as with an high-level dedicated UNIX server machine, featuring 2 Intel Xeon processors, 256 Gb of RAM and nearly 70 Tb of available hard-disk storage space.



Genomics & Transcriptomics

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Overview

The Genomic and Transcriptomic (GT) Unit at CAAD is aimed at producing and analyzing data in the genomic and transcriptomic field obtained with NGS and/or arrays, both for UPO researchers as well as external users.



Unit's staff supports users in designing, performing and interpreting high-throughput genomic and transcriptomic experiments for biomarker discovery and/or analyses of multifactorial and monogenic traits.

GT Unit operates tightly with Bioinformatics Unit at CAAD and offers an integrated pipeline which covers the entire process from the purification of nucleic acid from biosamples to the bioinformatic interpretation of the results. In addition, the Unit offers the interpretation of the genomic results and the preparation of the medical report, suitable for genetic counseling, to medical specialists, patients and researches.

Services:

This Unit offers:

- Laboratory and computational support for all the main type of high-throughput NGS sequencing analysis such as target sequencing of gene panels, whole exome sequencing, mRNA and microRNA- sequencing.
- Sanger sequencing as well as support to nucleic acid purification and quantification.
- Diagnostic tests using either validated genetic diagnostic tests as well as new genetic tests developed and validated within projects of the Institute as well as upon requests from external laboratories and partners.

These analyses are mainly based on a local implementation of state-of-the-art NGS sequencing pipelines. In addition to that, the Unit offers consultancy for non-standard analysis and support over long-term projects.

Equipments:

- Next Generation Sequencing Instruments: Illumina MiSeq System and NextSeq 550 Sequencing System
- Infinium Option Starter kit for the manual preparation and scanning of the Infinium BeadChip arrays such as Infinium[®] HumanCytoSNP-12, CytoSNP-850K, HumanKaryomap-12



BeadChips, for the identification of Copy Number Variant in human diseases, and the Methyl EPIC 850 array for whole genome human methylation assay of 850,000 CpGs

- SeqStudio Genetic Analyzer (ThermoFisher Scientific, Life Technologies) for Sanger sequencing and DNA fragment analysis for applications such as Microsatellite/tandem repeat sizing and genotyping, MLPA assay
- Real time PCR Bio-Rad CFX384 qPCR and Bio-Rad CFX96 qPCR Instrument, droplet digital PCR (ddPCR, QX200 Bio-Rad)
- Agilent TapeStation (TapeStation 4200), a system for sample Quality Control during NGS workflows
- Automatic workstation for liquid transfer (Tecan Freedom EVO100)

Next-Gen flow Cytometry & Sorting

Contact: annalisa.chiocchetti@uniupo.it

Overview

Next-Gen flow Cytometry & Sorting (NGCS) Unit is equipped with the novel BD FACSymphony[™] analyzer, that enables the simultaneous measurement of up to 29 different antigens of a single cell, and the BD FACSAria[™] Fusion sorter (12 colours).

Services:

The Symphony analyzer is an open service and can be operated by the users after training by the facility staff. The staff may support the users in experimental design and data analysis or supply an all-inclusive service.

Sorting is a restricted service that will be operated by facility staff only.

Equipments:

- BD FACSymphony[™] analyzer
- BD FACSAria[™] Fusion sorter

Metabolomics & Proteomics

Contact: marcello.manfredi@uniupo.it

Overview

The Unit Metabolomics & Proteomics (MP) is equipped with state of-art instrumentation to provide comprehensive analyses of proteins, small molecules, and lipids from different matrices (e.g.: cells, biological fluids, environment, food ...).

The laboratory is centered around two high-resolution mass spectrometers coupled to liquid chromatography and one bidimensional gas chromatography mass spectrometry.

If needed, the staff can develop new customized analytical methods and can perform data analysis, pathway analysis and omics data integration.

Services:

The Proteomics and Metabolomics Lab offers a range of different services

Proteomics:

- Global quantification of proteomes (label-free and label-based)
- Biomarkers discovery (immunodepletion of most abundant proteins)
- Immunoprecipitation analysis
- Identification and site-localization of selected post-translational modifications
- Identification of gel-separated, Coomassie-stained and WB proteins
- Protein fractionation with LC
- Data analysis for all the services listed above
- Storage of raw and processed MS data in publicly available repositories

Metabolomics:

- Global untargeted metabolomic profiling
- Targeted metabolomic profiling
- Lipidomic analysis
- Metabolite and lipid identification
- Comparative metabolic profiling (UPLC-MS-MS and GCxGC-MS)
- High throughput data analysis (including basic statistics)

Equipments:

The Unit is equipped with:

- Thermo Scientific Q Exactive Plus Mass Spectrometer + Vanquish UHPLC Systems
- Sciex Triple TOF 5600 plus + micro LC 200 eksigent or easy nanoLC Thermo
- LECO Pegasus BT 4D GCxGC-TOF (SPME and HS)
- Ultimate 3000 biocompatible + DAD

- Gel-proteomics (SERVA HPE blue Horizon, IEF Biorad, Chemidoc)
- FastPrep 24, Ultraturrax, point sonicator
- Positive pressure-96 for SPE
- Liquid handling for ELISA
- Bioinformatic analysis: IPA Ingenuity Pathway Analysis, Cytoscape, STRING, PANTHER, Metaboanalyst, etc.

Metagenomics

Contact: flavio.mignone@uniupo.it

Overview

The Metagenomics (META) Unit identifies the quality and quantity of the microorganisms present in an environment (microbiota) through the analysis of their genomes (microbiome) and transcriptomes (meta-transcriptomics).

The Unit works in synergy with other units to provide a broad range of analysis starting from "outof-the-box" first-level profiling of microbiota to high level study of meta-genomes/metatranscriptomess to highlight the functional capacity of the organisms.

Services:

Currently available services comprise the sequencing of the 16S rDNA gene to profile the microbiome community in a complex biological sample.

16S rDNA regions: V1-V2-V3 or V3-V4-V6

The service comprises:

- DNA extraction
- DNA Quality Control
- Library preparation
- Sequening
- Data analysis
- Report:
 - o Raw results: text file with abundances for any taxonomic level
 - Standard report: pdf file summarizing sequences statistics, alpha diversity, classification of top 10 abundances for each taxonomic level (table and chart), quality information about unassigned sequences.
 - Custom report: a custom pdf file with other information can be provided. It is also possible to add a personal logo and footer to the document.

For specific purposes, bacteria identification can be performed by MALDI TOF analysis (Biotyper app).

Protein Technologies

Contact: diego.cotella@uniupo.it

Overview

The Protein Technologies (PT) Unit provides expertise, tools and reagents for the custom production, purification, and analyses of recombinant proteins.

Services:

The Protein production service includes:

- in silico design (e.g. codon optimization)
- plasmid cloning
- expression into a prokaryotic host (E. coli) or mammalian cells (CHO)
- purification by affinity chromatography

The Biochemical assays service includes:

- receptor-ligand interaction (surface plasmon resonance)
- enzyme assay development
- inhibitor screening (medium throughput)
- functional assay in cellular models
- suspension array system for multiplex protein and nucleic acids analysis

The Unit offers:

• a set of vectors to express proteins in bacteria (E. coli) and mammalian cells CHO) cells (E. coli strains, CHO-S clones)

- reagents (recombinant enzymes for molecular biology, protein biochemistry and highthroughput screens)
- equipment (incubators, bioreactors, chromatography apparatus, surface plasmon resonance equipment)
- advice and protocols for protein purification and analysis
- full service (from the DNA in silico to the purified protein)
- assistance and a risk-free service: We do NOT charge if we cannot deliver the protein

Equipments:

The Unit is equipped with:

- Shaking incubators
- Benchtop bioreactors
- Automated Liquid Chromatography systems (Bio-Rad)
- Sonicator
- Ultracentrifuge
- Surface Plasmon Resonance (SPR) analyser (Biacore X100)
- Bio-Plex 200 Bio-Rad

In the context of this Unit, Valeria Caneparo (valeria.caneparo@uniupo.it), a technologist working at CAAD, offers assistance for SPR analysis with Biacore X100. Surface plasmon resonance (SPR) is a label-free, real- time technique capable of measuring binding affinities and kinetics for biomolecular interactions. SPR has become a key bio-sensing technology in the areas of biological research and medical sciences, and it allows the real-time detection and monitoring of biomolecular binding events. In a Biacore experiment, one of the interacting molecules (the ligand) is bound to the biosensor surface, whereas the other (the analyte) is delivered to the surface in a continuous flow through a microfluidic system. Binding of the analyte to the ligand is monitored on real-time by surface plasmon resonance, which detects the mass concentrations at the surface. The response

is directly proportional to the mass of analyte that bind to the surface. A range of different sensor chips is available for different application purposes.

The technology requires no labelling and provides quantitative information on kinetic parameters (association and dissociation rate constants, ka and kd), affinity constants (K_D) and stoichiometry. Applications include epitope mapping, molecular assembly and small molecule screening. Moreover, experiments can be performed using crude media (cell lysates, tissue extracts, sera and biological fluids) allowing, in combination with mass spectrometry, the identification of binding partners of a given molecular target immobilized on the biosensor.

In addition, Valeria Caneparo is also providing assistance with Droplet Digital PCR (ddPCR) experiment. The Bio-Rad QX200[™] Droplet Digital[™] PCR System is a technology that provides high-precision with absolute quantification of nucleic acid targets without a standard curve and without dependence on amplification efficiency. ddPCR affords scientists the ability to quantify template molecules that may be undetectable using traditional techniques. Researchers are utilizing ddPCR to quantify genomic alterations, such as copy number variation, detect rare sequences, and determine absolute quantification of target DNA copies without the need for a standard curve.

The system includes a Droplet Generator that generates 20,000 single nanoliter-sized droplets from one ddPCR reaction containing your target or background DNA. Samples are processed 8 at a time. Hydrolysis probes (Taqman) or EvaGreen (SYBR-like) chemistry can be used on the system. After PCR the samples are transferred to a QX 200 Droplet Reader where individual droplet intensities are read.

ddPCR Applications:

- Liquid Biopsy
- Cancer biomarker discovery
- Copy Number Variation (CNV)
- Rare Sequence Detection
- Gene Expression Analysis
- Single-Cell Analysis
- Pathogen Detection and Microbiome Analysis
- Next Generation Sequencing (NGS)