

# **RNA-seq Workshop**

An introductory course to RNA-seq

(Torino 26<sup>th</sup>-30<sup>st</sup> March 2018)

# Further Information

For more information, please contact the course organizer: Prof. Raffaele A. Calogero Bioinformatics and Genomics Unit, Molecular Biotechnology Center, Via Nizza 52, 10126, Torino Italy.

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# **Teaching Format**

This course will include a series of theoretical sessions followed by practical exercises. This course will utilize open-source software All software is embedded inSeqBox (www.seqbox.com). SeqBoxes will be provided by the B&Gu.

# Aims and Objectives

At the end of the course you will be able to:

- ✓ understand the importance of experimental design in order to ask sensible biological questions
- ✓ assess the quality of your data
- ✓ complete basic statistical tests on Next Generation Sequencing (NGS) data
- ✓ annotate and interpret your data and perform integration between gene-level expression and microRNA differential expression data
- ✓ understand some of the problems encountered when analyzing data
- ✓ acquire basic skills in R scripting

#### Audience

This course is suitable for biologists who are new to Next Generation Sequencing technology. Knowledge of statistics is not necessary prior to attending the course.

# Course Description

#### Tools for RNA-seq data analysis

The course is based on the use of Bioconductor open-source software solutions. However, R coding skill is not required. Furthermore, RNAseq and miRNAseq analyses will be also performed using the apps available in BaseSpace (https://basespace.illumina.com/home/index)

## Experimental design

This section of the course discusses several criteria and principles of experiment design as well as related problems. Questions such as how many replicates one needs to detect differential gene/microRNA expression or alternative splicing events are addressed.

#### Quality control

This section will focus on RNA-seq quality controls. Approaches to check the quality of raw data will be presented as well as approaches to identify sequencing bias. Approaches to experimental replicates will also be considered. All approaches will be practically tested on real data provided during the practical training sessions.

#### **Basic Statistics**

This part will provide the biologist with a general overview on issues closely related to RNA-seq data. The purpose is to give only as much information as needed to be able to make an informed choice during the subsequent data analysis. The aim of the training module is to put things in the perspective of someone who analyzes gene/exon-level RNA-seq data, rather than offer a full treatment of the respective statistical notions and techniques. No previous statistical knowledge is assumed.

Selecting differentially regulated genes/microRNAs

## Instructor Credentials Raffaele Calogero is

Associate Professor at Turin University and the P.I. of the Bioinformatics and Genomics unit. The Bioinformatics and Genomics unit (B&Gu) is a core facility to support researchers in multiplatform microarray/RNAseq experimental design, analysis and mining. Since 2002 he has led theoretical/practical training courses on microarray data analysis. Since 2010 he is part of the training team of the EMBL Whole transcriptome data analysis course (Heidelberg,DE)

Marco Beccuti is a researcher at Dept. of Computer Science, University of Torino. He has a degree and a PhD in Computer Sciences. He is involved in NGS tools development.

Francesca Cordero is a researcher at Dept. of Computer Science, University of Torino. He has a degree and a PhD in Computer Sciences. He is involved in NGS tools development.

Martina Olivero is a research technician of University of Torino.

Vladimir Benes is the head of the Genomics Core lab at EMBL Heidelberg. His lab assists researchers with functional genomics projects. He is strongly involved in teaching of methods applied to genomics and transcriptomics.

Nicola Cirenei is District Marketing Manager South Europe at Illumina

Organized by:

This portion presents several methods used to select differentially regulated genes/microRNAs in comparative experiments. The advantages and disadvantages of all methods are discussed in detail.

#### Selecting alternative splicing events

This portion presents approaches to identify alternative splicing events in a two groups experiment. The advantages and disadvantages of various methods are discussed in detail.

#### Single-cell RNAseq workflow

The field of single-cell genomics is advancing rapidly and is generating many new insights into complex biological systems, ranging from the diversity of microbial ecosystems to the genomics of human cancer. This module will briefly describe the differences existing between bulk RNAseq and singlecell sequencing. The module will provide a brief overview of the analysis of this type of data.

## Gene Ontology enrichment and data visualization

This session will focus on the extraction of biological knowledge from a set of differentially expressed genes using on-line tools like as EnrichR. Furthermore, the use of PCA and hierarchical clustering will be described. As visualization tool will be used MeV, a Java application designed to allow the analysis of high-throughput data to identify patterns of gene expression. R pills

Learning a scripting language might be frustrating for biologists. Thus for the first three days of the course we will have two session of R scripting training (30 mins theory/30 mins practice) one after breakfast and the other before dinner. The fourth day of course will be completely devoted to the use of R scripting in setting up a basic differential expression analysis using Bioconductor packages.

## Practical sessions

The course is structured to provide practical analysis skills to the students. Datasets will be provided by B&Gu. Data provided by the organizers are based on cell lines experiments.

## **Dates Times and Locations**

The RNA-seq workshop will last two and half days, in March 2017.

 Day 1
 26<sup>th</sup> March
 14:00 – 18:00

 Day 2
 27<sup>th</sup> March
 10:00 – 18:00

 Day 3
 28<sup>th</sup> March
 10:00-18:00

 Day 4
 29<sup>th</sup> March
 10:00-18:00

 20:30
 – 22:00 Social dinner sponsored by B&Gu

Day 5 30<sup>th</sup> March 9:00-15:00

## **Course Costs**

## The cost of the course is 500 Euros (max 20 persons)

A booklet with all presentations, coffee breaks, lunches and the social dinner are provided as part of the course.







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