



RNA-seq Workshop

An introductory course to RNA-seq

(Torino 1st-2nd March 2012)

Further Information

For more information, please contact the course organizer:
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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 and hardware 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
- ✓ perform normalization and reformatting procedures
- ✓ 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

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 since all the analyses are performed using oneChannelGUI, a graphical interface to Bioconductor tools, designed for life scientists who are not familiar with R language.

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.

Normalization and data reformatting

This section concentrates on data preparation issues. RNA-seq primary mapping tools will be described and their output will be reformatted to be uploaded in oneChannelGUI. Normalization and data reformatting will be applied to real data by the students.

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

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/RNA-seq experimental design, analysis and mining. Since 2002 he has led theoretical/practical training courses on microarray data analysis. Since 2008 he is part of the training team of the EMBL Whole transcriptome data analysis course (Heidelberg,DE)

Francesca Zolezzi has joined Singapore Immunology Network (SigN) as Principal Investigator Technologist for the Functional Genomics Platform Laboratory in April 2011. Before she was the Scientific Director of Genopolis, an Italian scientific public institution based in Milan with the aim to develop, integrate and disseminate Functional Genomics supporting national and international infrastructures.

Francesca Cordero is a researcher of B&Gu. She has a degree in Biological Sciences and a PhD in Informatics.

Andrea Acquaviva is Assistant Professor at the Department of Control and Computer Engineering of Politecnico di Torino (DAUIN - EDA Group).

Lukas Smink is Product Marketing Manager, Sequencing & Informatics - EMEA, Illumina

Marco Cappelletti is Sr. Manager Field Marketing - Europe at Illumina

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

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 group experiment. The advantages and disadvantages of all methods are discussed in detail.

Multiple testing

This section discusses issues related to the fact that RNA-seq interrogates a very large number of genes/exons simultaneously and its consequences regarding data analysis. This is a crucial aspect that changes the nature of the data analysis techniques used. Yet, this aspect is often neglected, although issues related to multiple testing can easily invalidate otherwise well-conceived experiments.

Biological interpretation

This session will deal with the relationship existing between microRNAs differential expression and their effects on putative target genes. The limits of bioinformatics identification of microRNAs gene-targets will be addressed as well as the improvement that can be obtained integrating gene expression data.

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 full days, in March 2012.

Day 1 1st March 8:30 – 18:00
20:30 – 22.:00 Social dinner sponsored by B&Gu and Illumina

Day 2 2nd March 9.30 – 16:30

Course Costs

Option 1:

The cost of the course is 500 Euros in case participants decide to use the hardware provided by the organizers. **(max 15 persons)**

Option 2:

The cost of the course is 400 Euros in case participant decides to use own hardware. Minimal requirement are Windows7 or Linux 64 bits or MAC OS 64 bits operating systems. At least 4 Gb RAM and at least i5 Intel processor or equivalent. Persons that decide for this option will be contacted to provide them support for installation of all required packages for exercises. **(max 5 persons)**

Both above options provide a booklet with all presentations, coffee breaks, lunches and the social dinner at Adriano Mesa Restaurant (via Principe Amedeo 57 I - 10060 Frossasco).

AGENDA

DAY ONE			
08:30	09:00	Registration + Breakfast	
09:00	09:15	Course Introduction	RAC
09:15	09:45	About RNA-seq Illumina technology (T)	MC
09:45	10:45	Minimal requirements for RNA-seq design	LS
10:45	11:30	Tips on sample prep.	FZ
11:30	12:00	Break	
12:00	13:00	QC and Bias detection (T)	RAC
13:00	14:00	Lunch	
14:00	15:00	Primary mapping (T)	FC
15:00	15:30	Tips on oneChannelGUI Installation (T)	RAC
15.30	16.30	Tools for Differential Expression, gene-level analysis DE (mRNA) (T)	RAC
16:30	17:00	Break	
17:00	18:30	Exercises on QC, RNA-seq data loading, gene-level differential expression (E)	RAC/FC
19:30		Getting the BUS to the restaurant	
20:30	23:30	Dinner in Frossaco	

DAY TWO			
09:30	10:30	Tools for isoforms identification(SpliceTrap vs Cufflinks) (T)	FC
10:30	11:30	Tools for fusion products identification (T)	AD
11:30	12:00	Break	
12:00	13:00	Pipe-line for short non-coding differential expression and miRNA targets prioritization (T)	RAC
13:00	14:00	Lunch	
14:00	16:00	Exercises on miRNA differential expression and miRNA target detection. (E)	RAC/RS/FC
16:00	16:30	Break and wrap up	
16:30		Departure	

(T: Theory; E: Exercises; RAC: Raffaele A. Calogero; FZ: Francesca Zolezzi; FC: Francesca Cordero; LS: Lukas Smink, MC: Marco Cappelletti, AD: Andrea Acquaviva)

Venue Details

Aula Seminari, Centro di Biotecnologie Molecolari (MBC), Via Nizza 52 Torino, 10126 Italy.

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Travel Information

By Air (Torino Airport):

Torino International Airport receives flights from all of Europe

(<http://www.aeroporto.torino.it/IT/voli/default.php>). A taxi is approximately 40 Euros from the terminals to MBC

(http://www.mbcunito.it/how_to_reach.php).

By train (Porta Nuova station):

Torino can be reached by train (<http://www.ferroviedellostatunito.it/>). Take underground, direction Lingotto, get out at Nizza station. MBC is 100 meters from the station.

By Car

See directions at:

http://www.mbcunito.it/how_to_reach.php

Meals and

Accommodation

Lunch and coffee breaks will be provided on all days for the duration of the course. In addition there will be a group dinner for all course attendees on 1st March 2012.

Participants will need to organize their own

accommodation. You will be provided with hotel suggestion after course registration..

How to Register

Course Registration

The course is limited to 20: 15 participants for option 1 and 5 participants for option 2.

Please complete the registration at

www.bioinformatica.unito.it/RNAseq.course.html before **10st February 2012**

Once you have received confirmation of the availability of your seat, you can:

- make a credit card payment through the BITS online payment form: www.bioinformatica.it/index.php?mod=bitpos
The 'Motivation' must be: RNAseq2012 <name surname> BITS member YES/NO
In case you are a BITS member you are eligible to a 10% discount, i.e. 450 or 360 Euros depending on the type of registration.
Take note of the code that is returned at the end of the procedure. This code is in the format BITS-NNNN, where NNNN is a four digit number
- pay the registration fee via bank transfer to the course registration account:
Code for Italy
BANCA SELLA SPA
Agenzia di Rosta , Via Rivoli, 63 (TO) Italy
IBAN: IT 78 Z 03268 31260 052843266940
International code:
BBAN: Z 03268 31260 052843266940

When you specify the reason of this credit card/money transfer, please indicate: **RNAseq2012**

Please, send a proof of credit card (i.e. BITS-NNNN code)/money transfer via email (raffaele.calogero@unito.it) or fax (+39 0116706487)

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