



## *Genética Molecular e Populacional*

### **Course: Regulation of Gene Expression**

**Date:** 27 -30 April 2015

**Location:** IBMC – Auditorium C

**Coordinator:** Alexandra Moreira

**Faculty and invited speakers:** Alexandra Moreira, Natalia Gromak, Ana Pombo, Franco Pagani, Nicholas Proudfoot.

#### **Objectives of the course:**

Regulation of gene expression at the RNA level depends on a panoply of processes and factors, including chromatin remodelling, transcription, pre-mRNA processing, *cis* regulatory elements, RNA binding proteins, miRNAs, lncRNAs and mRNA localization. In this course international experts in the area will present state-of-the-art research on the molecular basis of gene expression regulation and mRNA processing during the physiological events that occur in different types of cells and in different systems. Scientific challenges in the field and how they are experimentally addressed will be discussed with the students.

**Outline of the course:** Lectures, tutorials, lab meetings, research seminars, Mini-Symposium on 'Mechanisms of Gene Expression in Health and Disease'.

## Monday 27<sup>th</sup> – Alexandra Moreira

### Morning

1. Introduction
2. Lecture 1: Overview of transcription and RNA processing; integration of co-transcriptional events.
3. Lecture 2, research seminar: Mechanisms of mRNA 3' end formation in Eukaryotes.
  - a. Pre-mRNA cleavage and polyadenylation: Signals and molecular mechanisms; Regulation of mRNA 3' end formation
  - b. Poly(A) signals and diseases
  - c. Alternative polyadenylation

### Afternoon

4. Paper dissection:  
Pinto, PAB, Henriques, H, Freitas, MO, Martins, T, Domingues, RG, Wyrzykowska, PS, Coelho, PA, Carmo, AM, Sunkel, CE, Proudfoot, NJ and Moreira, A (2011) RNA polymerase II kinetics in *polo* polyadenylation signal selection, The EMBO Journal, 30: 2431–2444

### Suggested reading:

- 1) Lutz CS, Moreira A (2011) Alternative mRNA polyadenylation in eukaryotes: and effective regulator of gene expression. Wiley Interdisciplinary Reviews – RNA, 2 : 23-31
- 2) Moreira, A (2011) Integrating transcription kinetics with alternative polyadenylation and cell cycle control. Nucleus, 2(6)

## Tuesday 28<sup>th</sup> – Alexandra Moreira, Marta Oliveira & Natasha Gromak

### Morning- Alexandra Moreira

1. **10:00 - GR Lab meeting: Marta Oliveira**  
*The students will participate in the weekly lab meeting of the Gene Regulation group. GABBA students are expected to actively participate in the scientific discussions of the GR group.*

### Afternoon- Natasha Gromak

1. Lecture (Part 1): Principles of gene expression - Polymerase II CTD code and histone code.
2. Lecture (Part 2): miRNAs and their role in the regulation of gene expression

3. Paper dissection:

K Skourti-Stathaki, N. J. Proudfoot and N. Gromak (2011) Human senataxin resolves RNA/DNA hybrids formed at transcriptional pause sites to promote Xrn2-dependent termination. *Molecular Cell* **42**(6): 794-805

Suggested reading:

- 1) Groh M, Gromak N. (2014) Out of balance: R-loops in human disease. *PLoS Genet.* 18;10(9):e1004630. doi: 10.1371/journal.pgen.1004630.
- 2) Groh M, Lufino MM, Wade-Martins R, Gromak N. (2014) R-loops associated with triplet repeat expansions promote gene silencing in Friedreich ataxia and fragile X syndrome. *PLoS Genet.* 1;10(5):e1004318.
- 3) Gromak N, Dienstbier M, Macias S, Plass M, Eyraas E, Cáceres JF, Proudfoot NJ.(2013). Drosha regulates gene expression independently of RNA cleavage function. *Cell Rep.* 2013 Dec 26;5(6):1499-510. doi: 10.1016/j.celrep.2013.11.032. Epub 2013 Dec 19. Erratum in: *Cell Rep.* 2014 Jun 12;7(5):1753-4.

**Wednesday 29<sup>th</sup> – Franco Pagani, Isabel Pereira-Castro & Ana Pombo**

**Morning – Franco Pagani**

Lecture: pre mRNA splicing defects and therapeutic strategies

Suggested reading:

- 1) Dal Mas, A., Rogalska, M.E., Bussani, E. and Pagani, F. (2015) Improvement of SMN2 Pre-mRNA Processing Mediated by Exon-Specific U1 Small Nuclear RNA. *Am J Hum Genet*, **96**, 93-103.
- 2) Dal Mas, A., Fortugno, P., Donadon, I., Levati, L., Castiglia, D. and Pagani, F. (2015) Exon-Specific U1s Correct SPINK5 Exon 11 Skipping Caused by a Synonymous Substitution That Affects a Bi-Functional Splicing Regulatory Element. *Hum Mutat*, **10.1002/humu.22762**.
- 3) Fernandez Alanis, E., Pinotti, M., Dal Mas, A., Balestra, D., Cavallari, N., Rogalska, M.E., Bernardi, F. and Pagani, F. (2012) An exon-specific U1 small nuclear RNA (snRNA) strategy to correct splicing defects. *Hum Mol Genet*, **21**, 2389-2398.
- 4) Pagani, F. and Baralle, F.E. (2004) Genomic variants in exons and introns: identifying the splicing spoilers. *Nat Rev Genet*, **5**, 389-396.

**12:00 - IBMC Post-doc Seminar Series: Global analysis of alternative polyadenylation regulators of MCL1 transcripts - Isabel Pereira-Castro, Gene Regulation Group, IBMC**

## **Afternoon – Ana Pombo**

Lecture (Part 1): Interplay between Polycomb repression and co-transcriptional RNA processing in mouse ES cells?

Lecture (Part 2): Chromatin 'communities' associated with RNA polymerase II on chromatin

### Suggested reading:

- 1) Brookes E, de Santiago I, Hebenstreit D, Morris KJ, Carroll T, Xie SQ, Stock JK, Heidemann M, Eick D, Nozaki N, Kimura H, Ragoussis J, Teichmann SA, Pombo A (2012) Polycomb associates genome-wide with a specific RNA polymerase II variant, and regulates metabolic genes in ES cells. *Cell Stem Cell* 10, 157-70.
- 2) Tee WW, Shen SS, Oksuz O, Narendra V, Reinberg D (2014) Erk1/2 activity promotes chromatin features and RNAPII phosphorylation at developmental promoters in mouse ESCs. *Cell* 156, 678-90.
- 3) Stock JK, Giardrossi S, Casanova M, Brookes E, Vidal M, Koseki H, Brockdorff N, Fisher AM, Pombo A (2007) Ring1B-mediated ubiquitination of H2A restrains poised RNA polymerase II at bivalent genes in ES cells. *Nature Cell Biology* 9, 1428-1435.
- 4) Brookes E, Pombo A (2009) Modifications of RNA polymerase II are pivotal in regulating gene expression states. *EMBO Reports* 10, 1213-9.

## **Thursday 30<sup>th</sup> - Nick Proudfoot & Mini-Symposium**

### **Morning- Nick Proudfoot**

Lecture (Part 1): Genomic nascent transcriptional analyses (NET-seq): association of co-transcriptional RNA processing with different transcriptional stages.

Lecture (Part 2): Transcriptional termination and R-loop formation. Transcription termination associated with lncRNA that harbor pre-microRNA

### Suggested reading:

- 1) Takayuki Nojima, Toma Gomes, Ana R. F. Grosso, Hiroshi Kimura, Michael J. Dye, Somdutta Dhir, Maria Carmo-Fonseca\* and Nick J. Proudfoot\* (2015) Mammalian NET-seq analysis reveals genome-wide nascent transcription coupled to RNA processing. *Cell* In press April 21st
- 2) Konstantina Skourti-Stathaki, Kinga Kamieniarz-Gdula K and Nick J. Proudfoot\* (2014) R-loops induce repressive chromatin marks over mammalian gene terminators. *Nature*. 516: 436-439
- 3) Ashish Dhir, Somdutta Dhir, Nick J.Proudfoot\* and Catherine Jopling\* (2015) Microprocessor mediates transcriptional termination in long noncoding microRNA genes. *Nature Struct. Mol. Biol.* In press March

**Afternoon**

**GABBA/IBMC Mini-symposium  
"Mechanisms of gene expression in health and disease"  
IBMC**

13:50 **Opening Session**

14:00 **Genomic analysis of RNA polymerase II nascent transcription and coupled RNA processing**

Nick Proudfoot, University of Oxford

14:45 **Molecular mechanisms of human neurodegenerative diseases**

Natasha Gromak, University of Oxford

15:25 **Pre mRNA splicing correction using Exon Specific U1 snRNP**

Franco Pagani, Human Molecular Genetics, International Centre for Genetic Engineering and Biotechnology, Trieste

16:05 **COFFEE BREAK**

16:20 **Polycomb repression acts through destabilization of nascent transcripts**

Ana Pombo, Berlin Institute for Medical Systems Biology, Max Delbrück Center for Molecular Medicine, Berlin

17:00 **Alternative modes of gene expression**

Alexandra Moreira, IBMC - University of Porto

**Course closing session and evaluation**

**6:30pm - Drinks with the students**

**Teachers contact list:**

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