Genética Molecular e Populacional

Course: Regulation of Gene Expression

Date: 9-13 May 2016
Location: i3S, Room B
Coordinator: Alexandra Moreira
Faculty and invited speakers: Alexandra Moreira, Natalia Gromak, Isabel Pereira-Castro, Jaime Freitas.

Objectives:
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, mRNA localization and local translation. In this course international 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 will be presented and discussed. Scientific challenges in the field and how they are experimentally addressed will be discussed with the students.

Outline of the course: Lectures, tutorials, paper discussions, research seminars, projects developed and presentations by the students.

Monday 9th – Alexandra Moreira & Natalia Gromak

Morning - Alexandra Moreira

1. Introduction; distribution of projects to be developed by the students.
2. *Lecture 1*: Overview of transcription and RNA processing. The transcription cycle: initiation, elongation, termination. The mRNA factory model.
3. *Lecture 2:* Integration of co-transcriptional events. RNA processing. RNA binding proteins. The complex life of RNA.

Afternoon - Natalia 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. Overall discussion with students' participation on integration of various processes of gene expression

Tuesday 10th – Natalia Gromak & Alexandra Moreira

Morning - Natalia Gromak

1. *Lecture*: Unusual RNA/DNA structures and their function in health (part 1) and disease (part 2).
2. *Paper dissection*:

   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.

Suggested reading:

1) 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
2) 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.
3) Gromak N, Dienstbier M, Macias S, Plass M, Eyras 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.

Afternoon - Alexandra Moreira

1. *Lecture 1*: Splicing; alternative splicing: biological relevance and regulation.
2. *Lecture 2:* Alternative splicing & disease; RNA therapeutics.
3. *Research seminar and paper dissection*:

   Glória, V, Martins de Araújo, M, Leal, R, de Almeida, SF, Carmo, AM and Moreira, A (2014) T cell activation regulates CD6 alternative splicing by transcription dynamics and SRSF1, J Immunol, 193(1): 391-399

Suggested reading:

1) Naftelberg S, Schor IE, Ast G, Kornblihtt AR (2015) Regulation of alternative splicing through coupling with transcription and chromatin structure. Annu Rev Biochem. 84:165-98.
2) Matlin AJ, Clark F, Smith CW (2005) Understanding alternative splicing: towards a cellular code. Nat Rev Mol Cell Biol. 6(5):386-98
3) Cooper TA, Wan L, Dreyfuss G. (2009) RNA and disease. Cell, 136(4):777-93.

Wednesday 11th – Alexandra Moreira & Jaime Freitas

Morning - Alexandra Moreira

1. *Lecture:* 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, function and regulation.
   d. The 3’UTR: a platform for gene expression regulation.
2. *Research seminar and 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) Mayr C. (2016) Evolution and Biological Roles of Alternative 3'UTRs. Trends Cell Biol. 26(3):227-37.
2) Lutz CS, Moreira A (2011) Alternative mRNA polyadenylation in eukaryotes: and effective regulator of gene expression. Wiley Interdisciplinary Reviews – RNA, 2 : 23-31

3) Shi Y, Manley JL. (2015) The end of the message: multiple protein-RNA interactions define the mRNA polyadenylation site. Genes Dev. 2015 May 1;29(9):889-97.

4) Curinha, A, Braz, SO, Pereira-Castro, I, Cruz, A and Moreira, A (2014) Implications of polyadenylation in health and disease, Nucleus, 5(6): 508-519

Afternoon - Jaime Freitas

1. *Research seminar (discussion of ongoing projects)*:

   a) RNA Polymerase II elongation rate and polyadenylation signal selection.

   Analysis of alternative polyadenylation by 3' Region Extraction and Deep Sequencing (3'READS).

   b) Heph/PTB and Elav/HuR are recruited to *polo* upstream sequence element that modulates alternative polyadenylation

   Characterization of an RNA sequence element and protein factors involved in *polo* alternative polyadenylation.

Suggested reading:

1) Fong, N., Kim, H., Zhou, Y., Ji, X., Qiu, J., Saldi, T., et al. (2014). Pre-mRNA splicing is facilitated by an optimal RNA polymerase II elongation rate. *Genes & Development*, *28*(23), 2663–2676.

2) Danckwardt, S., Kaufmann, I., Gentzel, M., Foerstner, K. U., Gantzert, A.-S., Gehring, N. H., et al. (2007). Splicing factors stimulate polyadenylation via USEs at non-canonical 3 ' end formation signals. Embo Journal, 26(11), 2658–2669.

Thursday 12th – Isabel Pereira-Castro & Alexandra Moreira

Morning - Isabel Pereira-Castro

1. *Research seminar* - Dissecting the molecular mechanisms controlling alternative polyadenylation in human T cells.
2. *Paper dissection*:

   Domingues RG, Lago-Baldaia I, Pereira-Castro I, Fachini JM, Oliveira L, Drpic D, Lopes N, Henriques T, Neilson JR, Carmo AM, Moreira A (2016) CD5 expression is regulated during human T-cell activation by alternative polyadenylation, PTBP1 and miR-204. Eur J Immunol. doi: 10.1002/eji.201545663. [Epub ahead of print]

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) Sandberg R, Neilson JR, Sarma A, Sharp PA, Burge CB (2008) Proliferating cells express mRNAs with shortened 3' untranslated regions and fewer microRNA target sites. Science, 320(5883):1643-7.
3) Baillat D, Wagner EJ (2015) Integrator: surprisingly diverse functions in gene expression. Trends Biochem Sci; 40(5):257-64.

Afternoon - Alexandra Moreira

1. *Lecture* – The role of RNA binding proteins and alternative polyadenylation in mRNA localization and local translation.
2. *Research seminars and paper dissection:*
   a) Braz *et al.* Alternative Polyadenylation of Rho GTPases: a gene and cell specific process, *in preparation*.
   b) Cruz *et al*, YBX1 function in oligodendrocyte differentiation and myelination, *in preparation*.

Suggested reading:

1) Kelsey C. Martin and Anne Ephrussi (2009) mRNA Localization: Gene Expression in the Spatial Dimension. Cell. 136(4): 719.
2) Hosung Jung, Christos G. Gkogkas, Nahum Sonenberg, Christine E. Holt (2014) Remote Control of Gene Function by Local Translation. Cell 157: 26–40

Friday 13th – Students projects

Morning - students will finish their projects under teacher's supervision if required.

Afternoon - students will present their projects and the course will be evaluated.

Faculty contact list:

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