Transcriptomic screen for DIS3 and DIS3L1 exosome subunits-associated functional networks in colorectal cancer

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Background

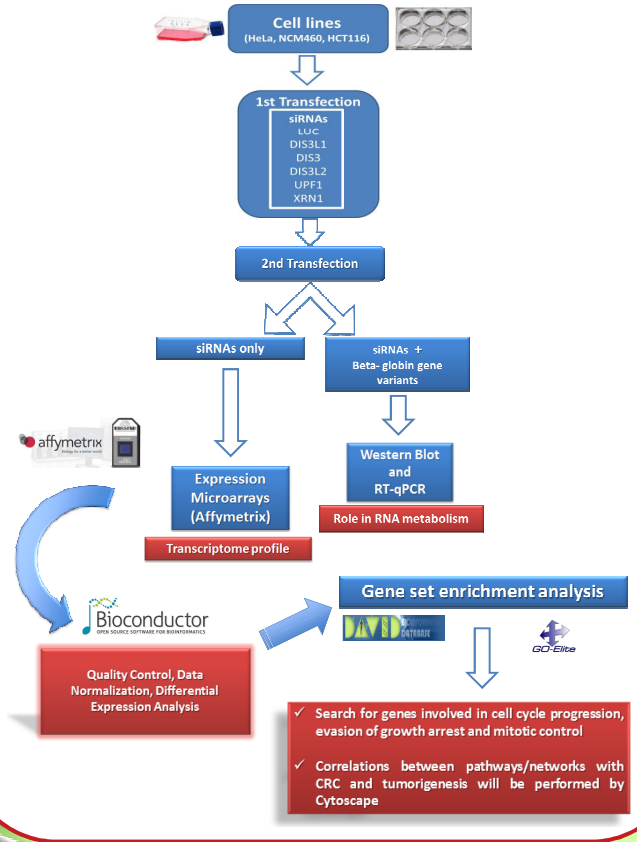
The final step of cytoplasmic mRNA degradation proceeds in either a 5'-3' direction, catalyzed by XRN1, or in a 3'-5' direction catalyzed by the exosome. In yeast, DIS3/Rrp44 protein is the catalytic subunit of the exosome. In humans, there are three known paralogues of this enzyme: DIS3, DIS3L1, and DIS3L2. Important findings over the last years have shed a new light onto the mechanistic details of RNA degradation by these exoribonucleases. In addition, it has been shown that they are involved in growth, mitotic control and important human diseases, including cancer. For example, DIS3L2 inactivation was associated with mitotic abnormalities and altered expression of mitotic checkpoint proteins (Astuti et al., 2012). In another study, DIS3 was found to be highly expressed in colorectal cancer (CRC), suggesting an oncogenic function (Camps et al., 2013).

A major challenge in systems biology is to reveal the cellular networks that give rise to specific phenotypes (Lan et al., 2013). In this project, we aim to analyze how DIS3 and DIS3L1 regulate the human transcriptome, and how their functional interactions modulate the transcriptional reprogramming of colorectal cancer cells.

Aims

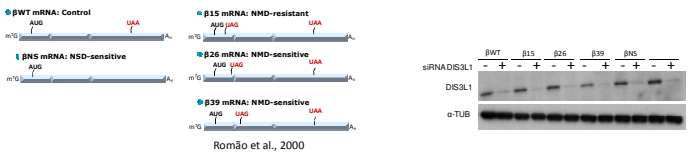
1. Investigate whether DIS3 and DIS3L1 are involved in the normal mRNA decay, as well as in the mRNA surveillance mechanisms of NMD and NSD and in the regulation of natural NMD targets, in HeLa, NCM460 (normal colon mucosa) and HCT116 (CRC) cell lines.
2. Characterize the DIS3L1 mRNA targets by DIS3L1 siRNA-mediated knockdown coupled to microarray profiling assays in NCM460 and HCT116 cells.
3. Elucidate new cellular pathways regulated by DIS3L1 and/or by their targets, as well as how they can be involved in CRC.
4. Reveal novel functional networks through which the exosome modulates the eukaryotic transcriptome.

Workflow

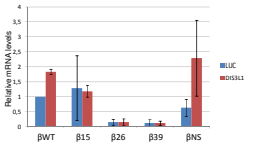


Results

What are the mRNA degradation mechanisms in which DIS3L1 is involved?

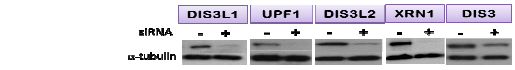


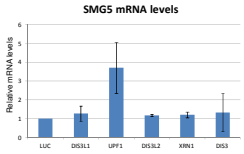
Romão et al., 2000



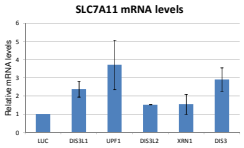
DIS3L1 seems to be involved in normal mRNA turnover and NSD, but not in NMD.

Do DIS3L1, DIS3 and DIS3L2 control mRNA levels of natural NMD targets?

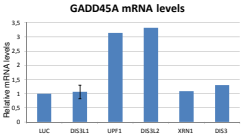




UPF1 is involved in the modulation of SMG5 mRNA levels whereas none of the others seem to be involved.



UPF1, DIS3L1 and DIS3 modulate SLC7A11 mRNA levels.



UPF1 and DIS3L2 modulate GADD45A mRNA levels.

The catalytic subunits of the exosome and DIS3L2 might have some target specificity! (depending on transcripts features??)

Timeline

Time	Year 1 (2014)		Year 2 (2015)		Year 3 (2016)		Year 4 (2017)	
	Months 1-6	Months 7-12	Months 13-18	Months 19-24	Months 25-30	Months 31-36	Months 37-42	Months 43-48
PhD courses								
Objective 1								
Objective 2								
Objective 3								
Objective 4								

References

Astuti D, Morris MR, Cooper WN, Staals RH, Wake NC, Fews GA, Gill H, Gentle D, Shuib S, Ricketts CJ, et al. Germline mutations in DIS3L2 cause the Perlman syndrome of overgrowth and Wilms tumor susceptibility. Nat Genet 2012, 44:277–284.

Camps J, Pitt JJ, Emons G, Hummon AB, Case CM, Grade M, Jones TL, Nguyen QT, Ghadimi BM, Beissbarth T, Difilippantonio MJ, Caplen NJ, Ried T. Genetic amplification of the NOTCH modulator LNX2 upregulates the WNT/β-catenin pathway in colorectalcancer. Cancer Res 2013, 73(6):2003-13.

Lan A, Ziv-Ukelson M, Yeger-Lotem E. A context-sensitive framework for the analysis of human signalling pathways in molecular interaction networks. Bioinformatics 2013, 29: 210-216.

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