Contribution of lncRNAs in Establishment of HIV Latency in Central Memory CD4 T Cells

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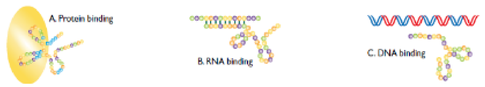
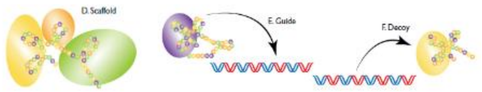
Introduction

- **HIV-host transcriptome studies** focused on **protein coding genes**
- **Majority** of transcribed sequences originate from **non-coding RNA**
- **lncRNA** are largely **unexplored** in **HIV infection** / replication cycle
- lncRNAs can bind all biomolecules in the cell



- Found in **human diseases**, **cell type/process specific**
- **Control transcription/translational** processes in the cell



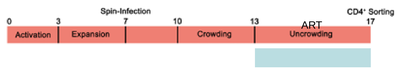
- Ideal **targets** for **viruses** to quickly **reshape gene expression**

Goal

- Study HIV latency mechanisms and explore **new treatment** strategies
- **Primary cell model** for main reservoir of HIV: CD4 T cells: Planelles
- Focus on **lncRNAs** in HIV latency and cure research

Methods

- **Planelles Model**: Primary HIV latency infection model in **central memory CD4 T cells**

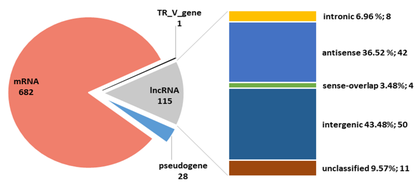


- **Flow cytometry** readout latently infected cells (4 replicates) of p24 HIV antigen after cell stimulation with antiCD3/CD28 and CD4 sorting
  - Latently infected cells: 2.12%-3.66%
- **Differential expression (DE)** between uninfected (UI) and latenty infected (LI) cells (total RNAseq, 4 biological replicates)
- **Bioinformatic** analysis
  - mRNA pathway analysis
  - lncRNA guilt-by-association analysis

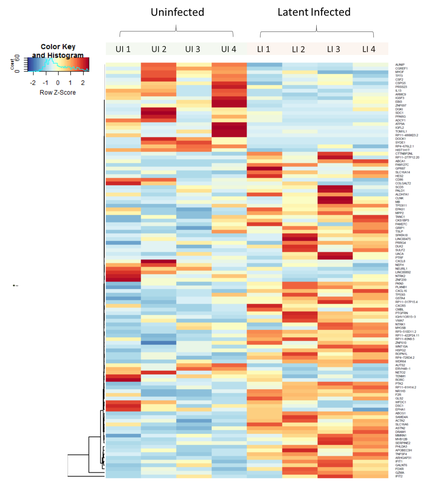
Results

Overview Differential Expression

- Biotype overview of 826 DE genes between UI and LI cells (FDR<0.05, adjusted p-value)



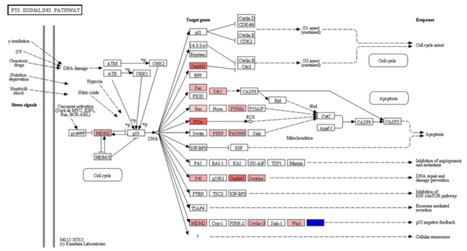
- Heatmap representation of top 100 DE genes between UI and LI cells



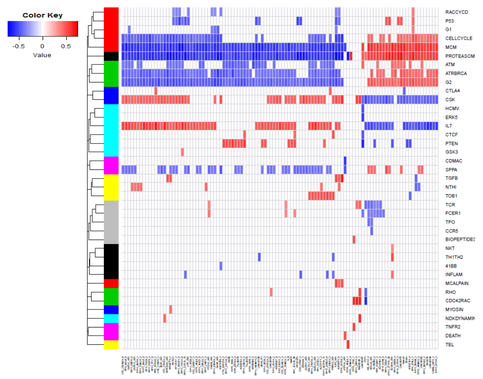
lncRNAs account for 115 DE genes in latently infected CD4 T cells

Pathway analysis mRNA/lncRNA

- p53 pathway enriched in mRNAs (1)



- Biological pathways for lncRNAs (guilt-by-association, enrichment scores with adjusted p-values < 0.05)



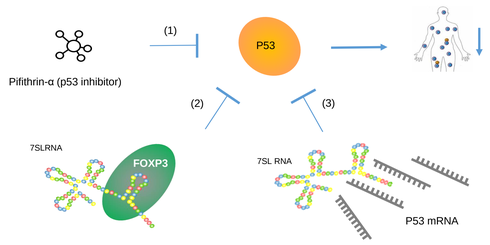
p53 pathway overlap with mRNAs
Other pathways: proteasome, IL-7, cell cycle

- mRNAs show p53 pathway enrichment
- lncRNAs could be linked to biological pathways with possible roles in HIV latency

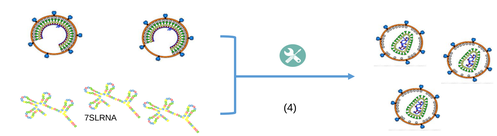
7SLRNA as potential new target

A) 7SLRNA was **upregulated** in latently infected cells and **negatively linked** to the **p53 pathway**

B) 7SLRNA may function as **p53 inhibitor** and mimic HIV latency block by pifithrin-α



C) 7SLRNA is **build into HIV** particles



D) 7SLRNA **interacts** with APOBEC3G (A3G) and is necessary for **antiviral A3G-response**

7SLRNA may be a new potential target in HIV latency cure strategies

Future Perspectives

- **Knockdown validation** of lncRNAs (locked nucleic acid based anti-sense oligonucleotides, Crispr Interference) in cell lines and CD4 T cells
- **Overlap** DE data of **different primary T cell models of HIV latency** (e.g. Spina model) to identify lncRNAs consistently dysregulated in latency
- **RNA pulldown** of lncRNA to identify **interaction partners**

References

(1) White et al. Plos Pathog 2016 12(11):e1006026.
(2) Yang et al. Biochem Biophys Res Commun 2016 472(3):432-6.
(3) Abdelmohsen et al. Nucleic Acids Res 2014 42(15):10099-111.
(4) Houzet et al. Nucleic Acids Res 2007 35(8):2695-704.
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