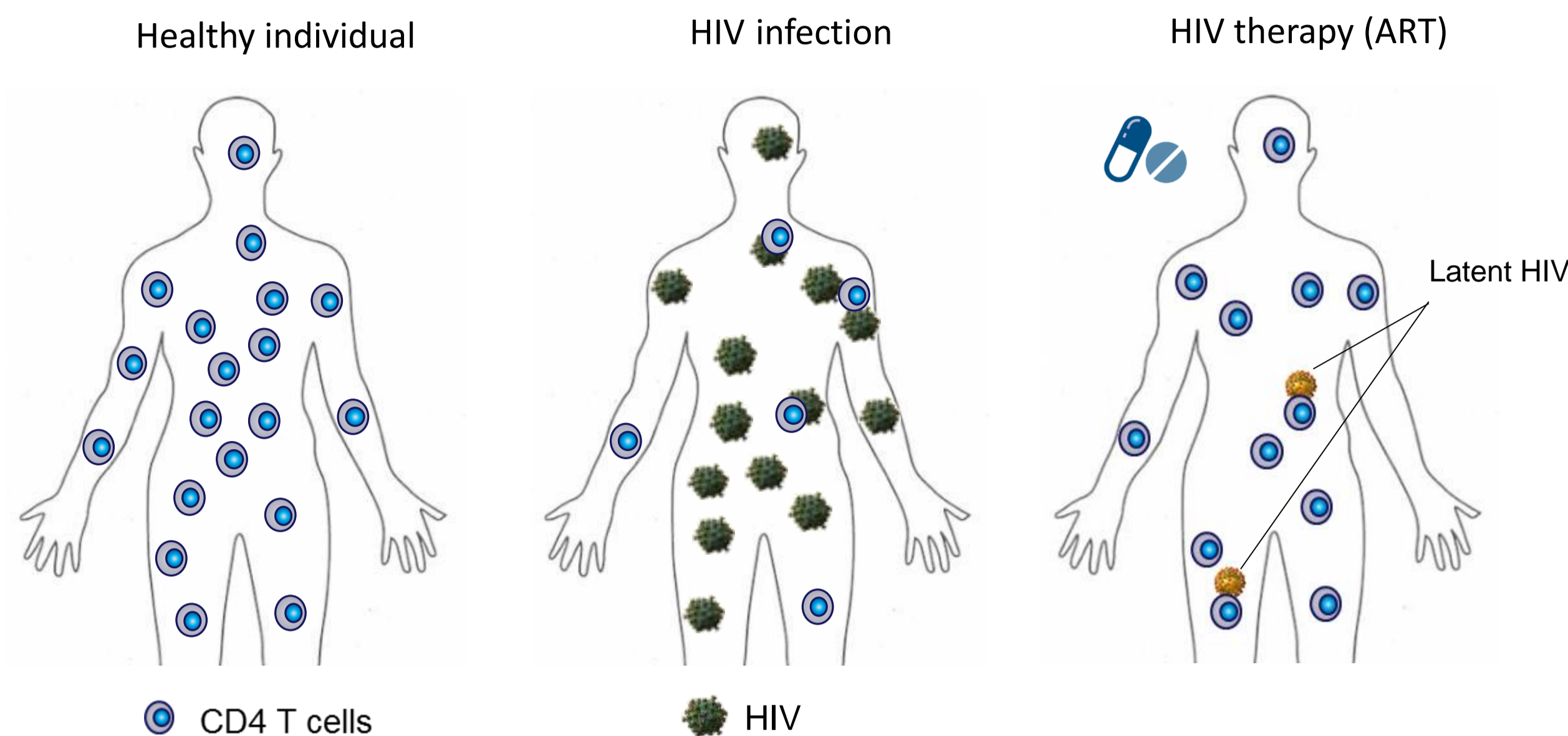


# lncRNAs and latent HIV - search for novel targets for latency reversal

Wim Trypsteen<sup>1</sup>, CH White<sup>2</sup>, A Mukim<sup>3</sup>, CA Spina<sup>3,4</sup>, W De Spiegelaere<sup>5</sup>, S Lefever<sup>6</sup>, CH Woelk<sup>2</sup>, A Bosque<sup>7</sup>,  
L Vandekerckhove<sup>1\*</sup>, N Beliakova-Bethell<sup>3,4\*</sup>

<sup>1</sup>HIV Cure Research Center, Ghent University, Belgium. <sup>2</sup>Faculty of Medicine, University of Southampton, UK. <sup>3</sup>San Diego VA Medical Center and Veterans Medical Research Foundation, USA. <sup>4</sup>University of California San Diego, USA. <sup>5</sup>Department of Morphology, Ghent University, Belgium. <sup>6</sup>Center Medical Genetics, Ghent University, Belgium. <sup>7</sup>University of Utah School of Medicine, USA. \*Contributed equally.

## Introduction



## Problem

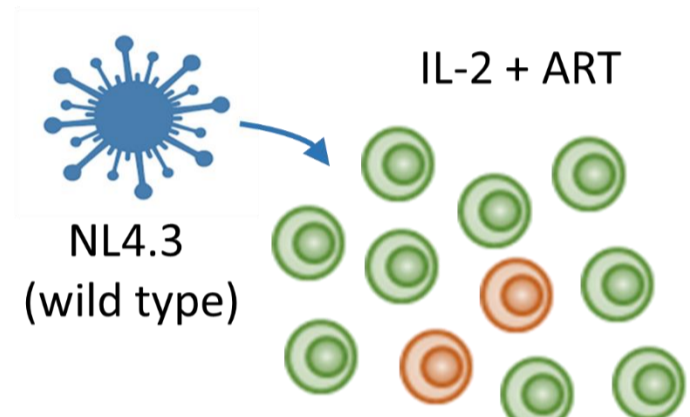
- Worldwide **35 million** people are **HIV-infected**
- HIV infection evolved from a deadly to a **chronic disease**
- Current treatment can suppress HIV but **not offer a cure**
- Condemned to **lifelong treatment** due to a **latent reservoir**

## Goal

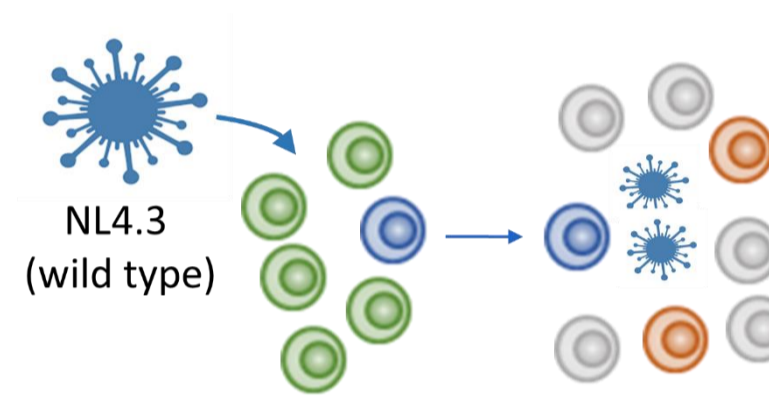
- Study HIV latency & explore **new treatment strategies**
- Primary cell models** for main reservoir of HIV: **CD4 T cells**
- Focus on **lncRNAs** in HIV latency and cure research

## Methods

### Cultured T<sub>CM</sub> latency model



- Direct infection of activated cells
- Differentiated CD4 T memory cells
- Return to resting state (latent)
- Latent cell fraction: 5-10%



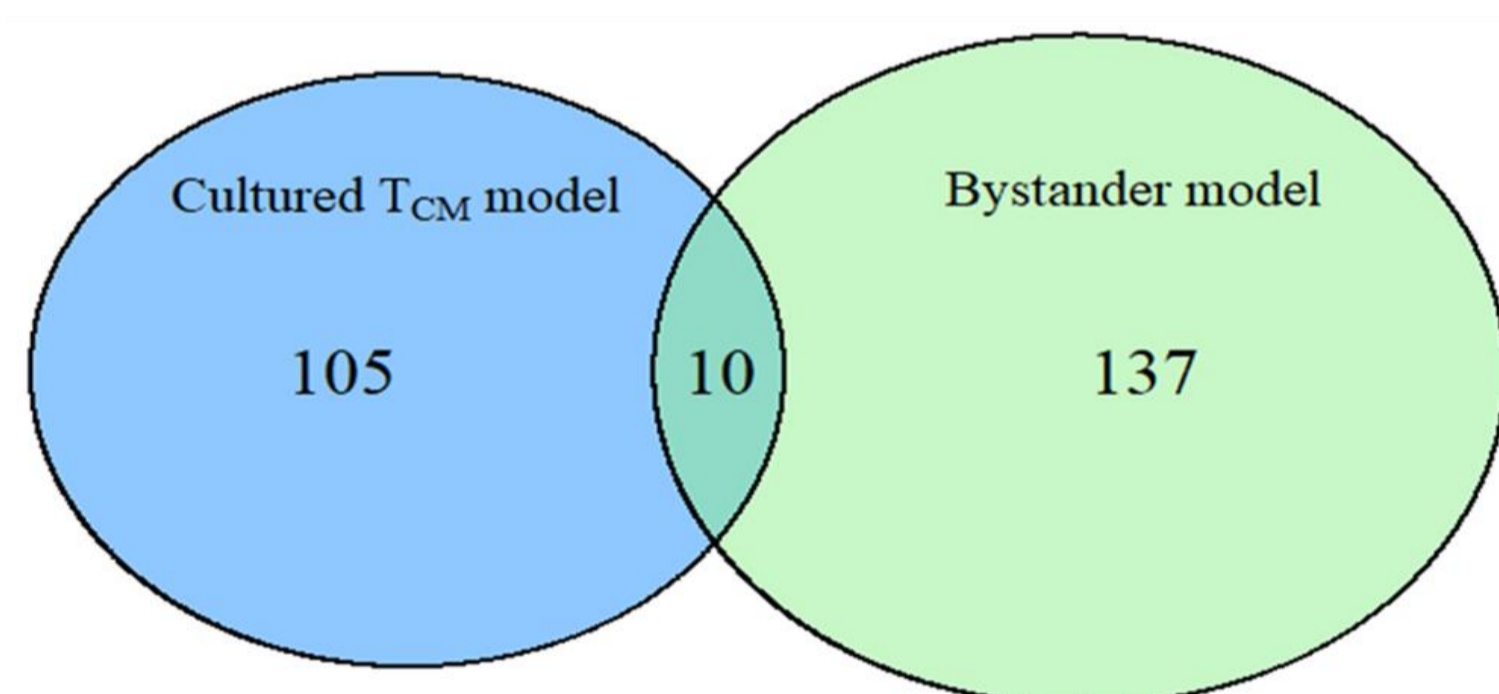
### Bystander latency model

- Two step infection: cell spreading
- Primary CD4 T cell pool
- Resting cells
- Latent cell fraction: 5-10%

- lncRNA discovery via total RNAseq, ribodepleted (4 biological replicates per model, Illumina Hiseq 2500, 30-50M reads per sample)
- Downstream analysis: Differential expression, ddPCR validation, pathway analysis and T cell subset analysis

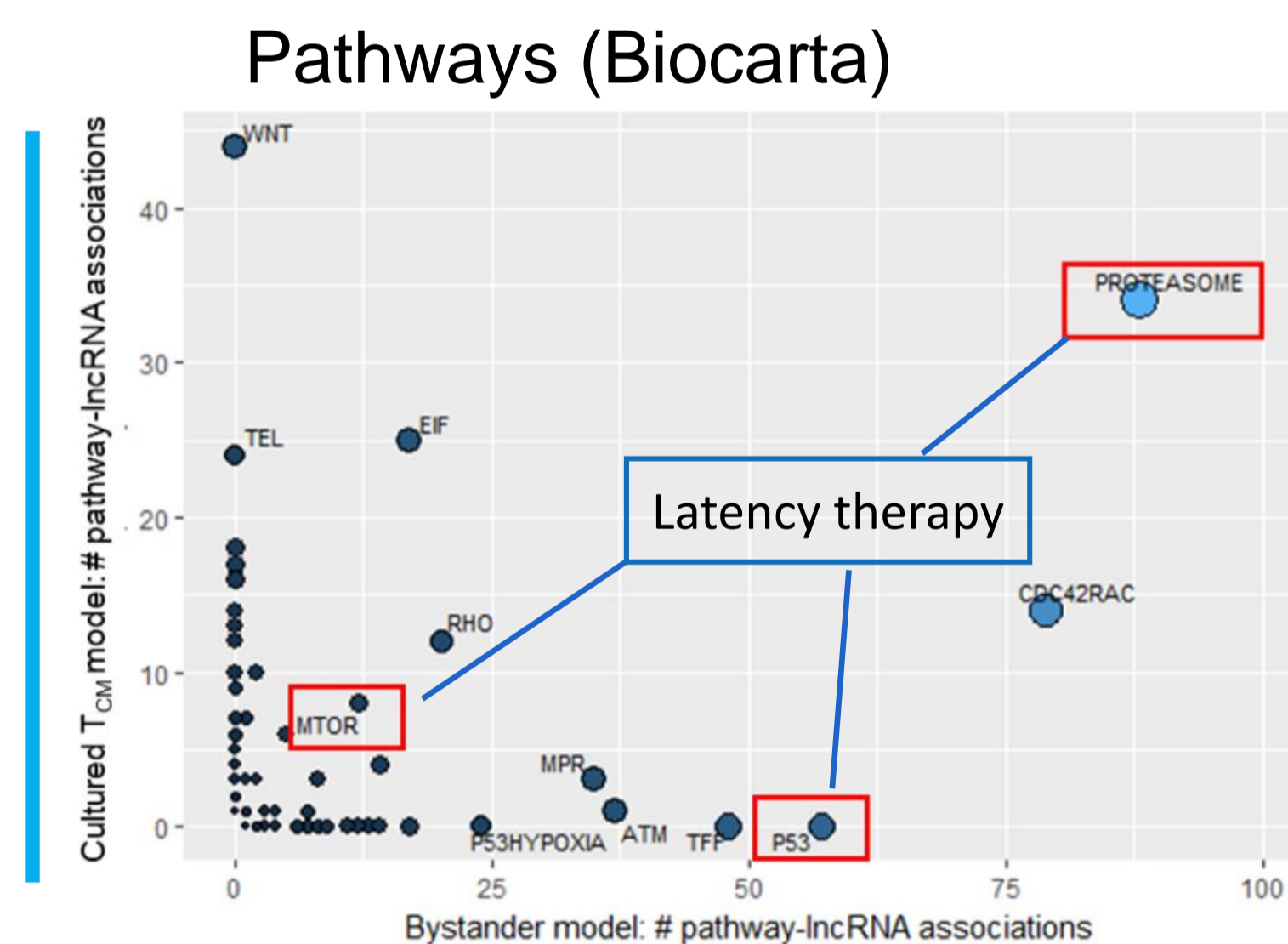
## Results

### 1 Differential Expression: lncRNAs

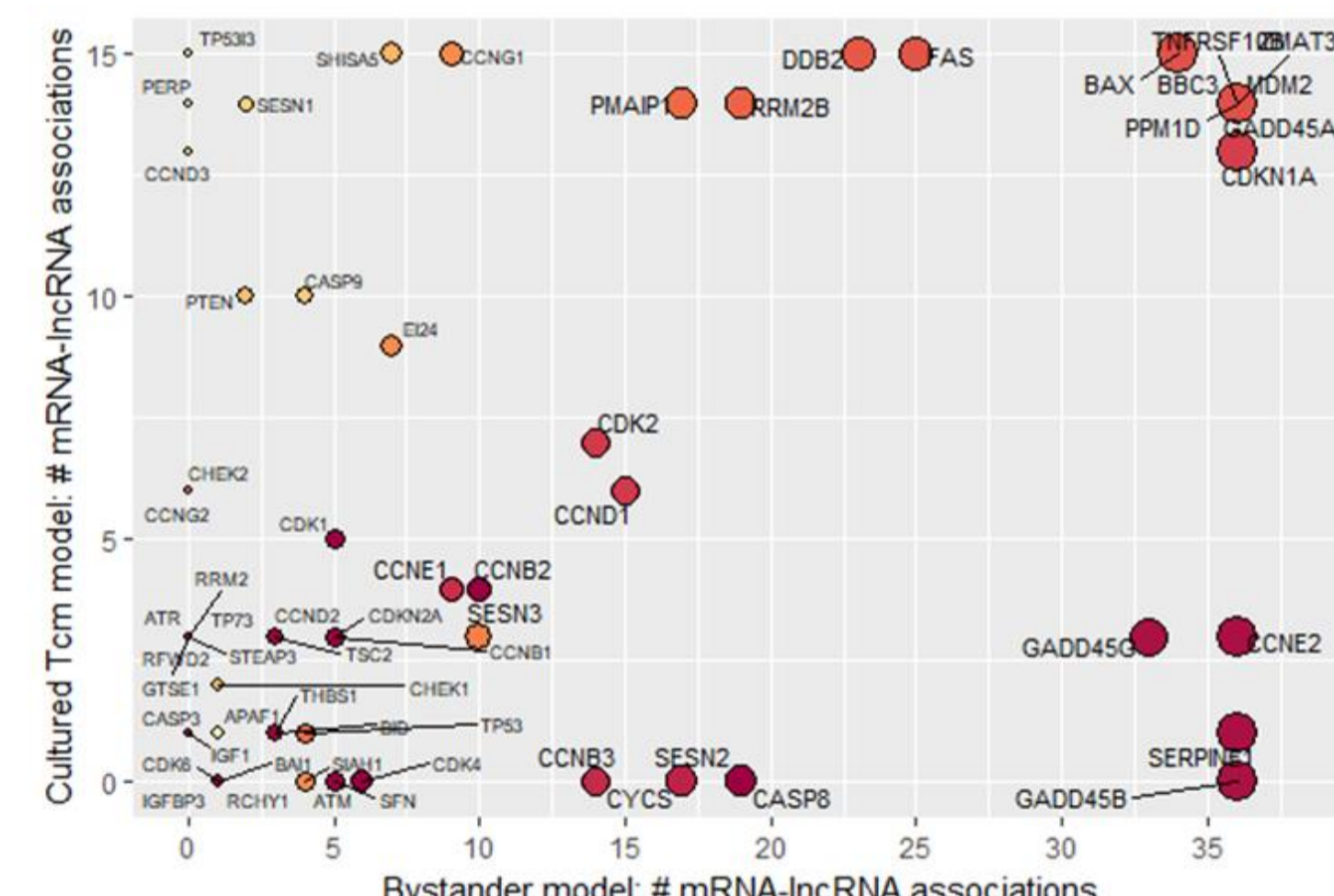


Digital PCR validated lncRNAs  
RP11-347C18.3, RP11-539L10.2, PVT1

### 2 Pathway analysis: Guilt-by-association

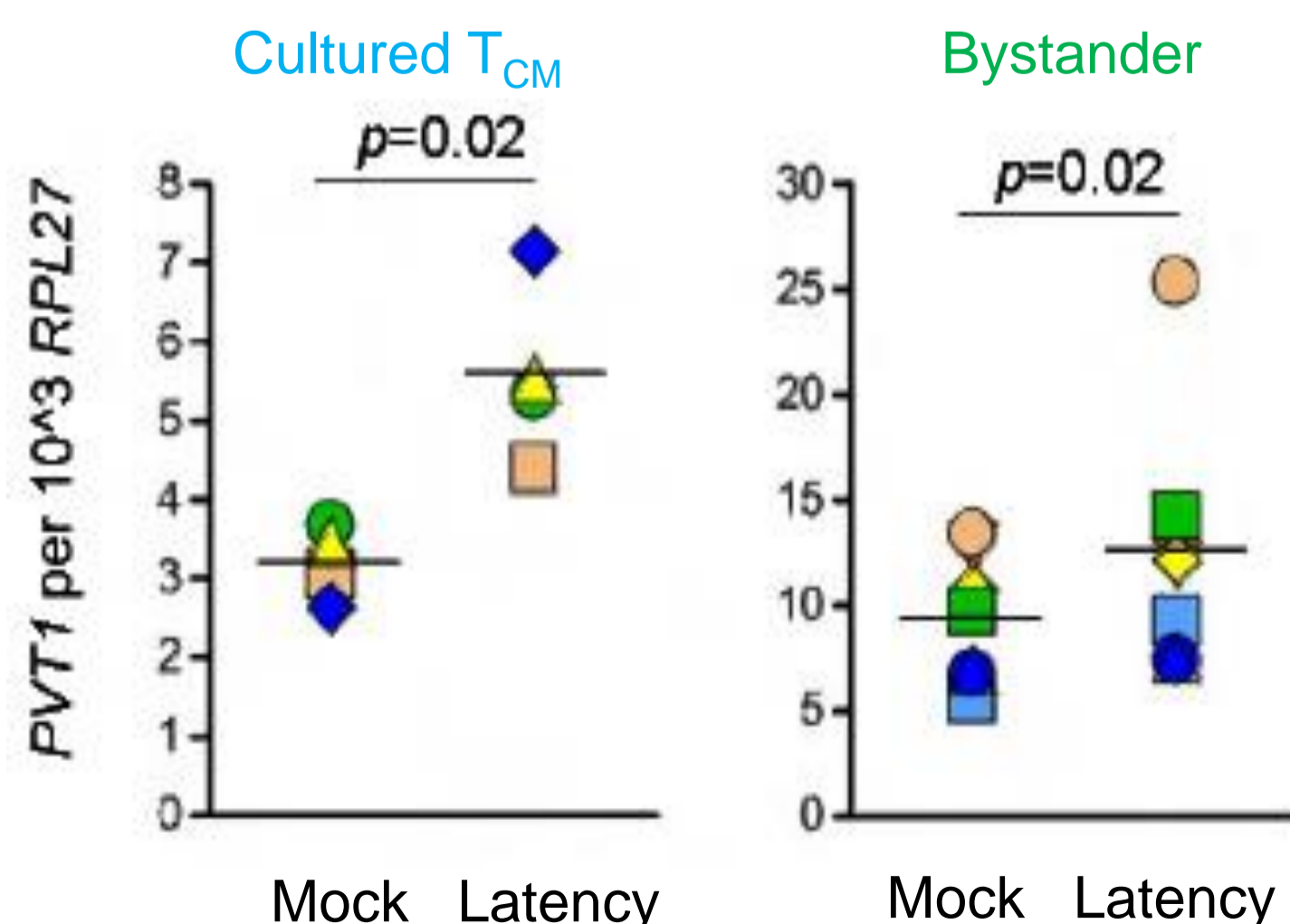


### P53 Detailed View

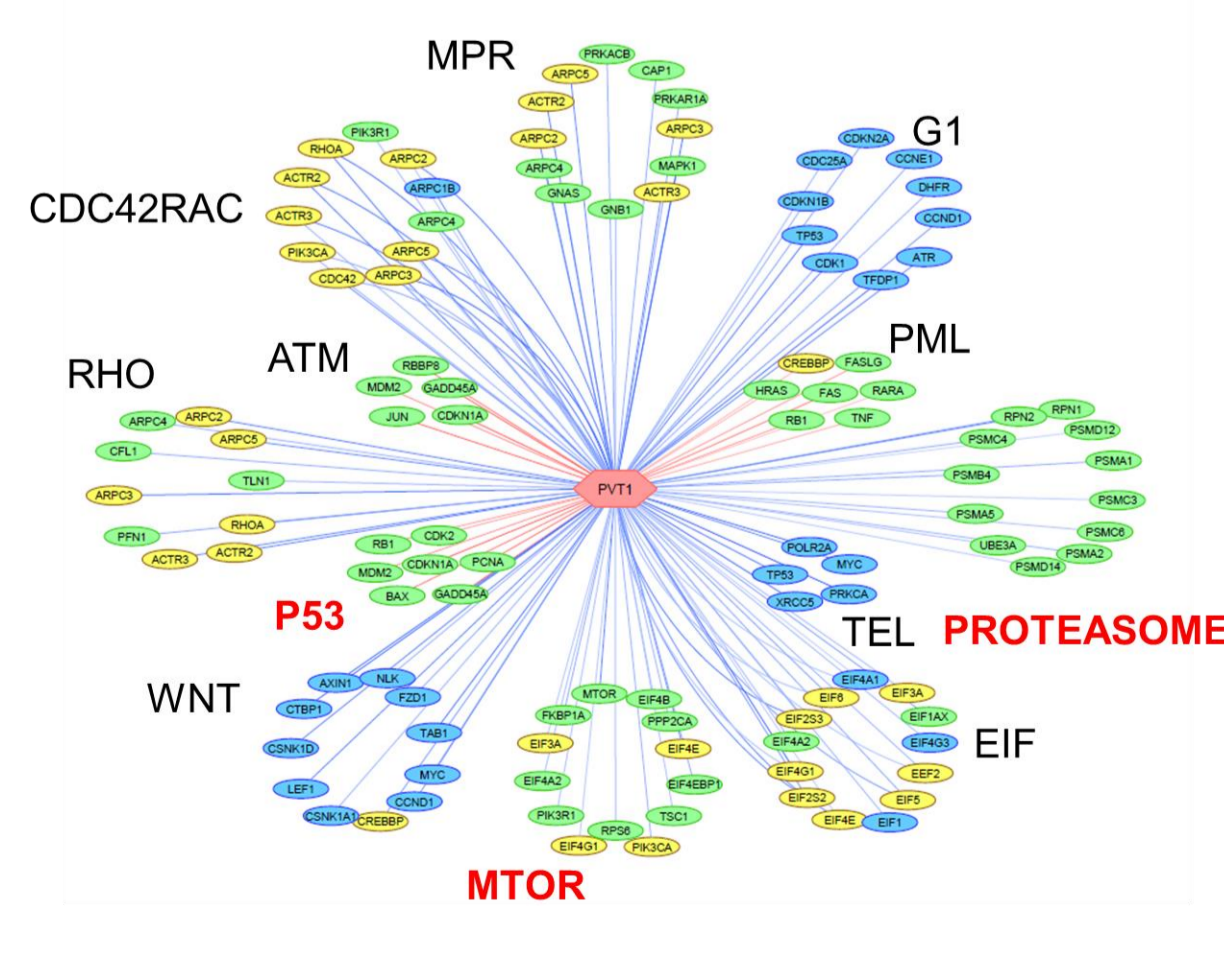


### 3 Focus on PVT1

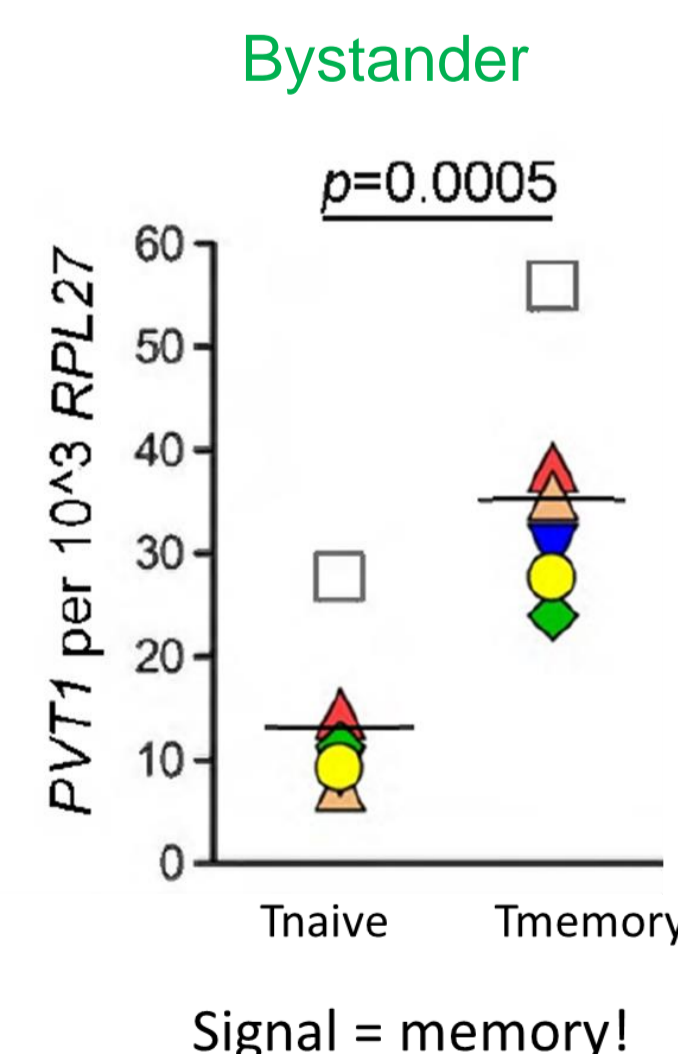
#### Digital PCR validation



#### Pathways associated



#### CD4 T cell subsets



## Conclusion

- lncRNA contribute to HIV latency
- lncRNA linked to **pathways** for possible therapy
- PVT1 is prioritized candidate** for further research
- Functional validation with **targeted knockdown studies** are required