# *Interactome of HIV proteins and their host RNA interaction partners*

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## BACKGROUND

The HIV genome encodes a limited set of proteins, depending heavily on the exploitation of host cell molecules to complete its viral life cycle. Recent findings show that HIV hijacks **cellular (non-coding) RNA molecules to aid in these crucial viral replication processes**. Therefore, this study aims to systematically determine this new layer of physical interactions of each of the 18 HIV proteins with host RNA molecules.

## METHODS

- HIV protein immunoprecipitation (with anti-FLAG Ab) followed by RNA sequencing (**RIPseq**) was performed (triplicate) in 18 Jurkat cell lines each expressing a single FLAG/STREP tagged HIV protein.
- Stranded total RNA library preparation and sequencing (50M reads/sample, Illumina NextSeq).
- After mapping (STAR), DEseq reveals enriched interaction partners of the HIV protein compared to background controls (mouse IgG antibody and FLAG-tagged GFP RIPseq).
- For a subset of 6 HIV proteins also streptavidin-based RIPseq was performed.

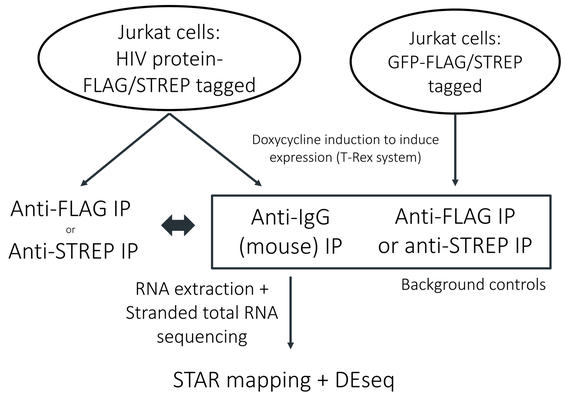


**Figure 1.** Schematic overview of the RNA Immunoprecipitation Sequencing (RIPseq) workflow performed for all 18 HIV proteins (all conditions in triplicate).

## RESULTS

- The identified interactome comprises a total set of 1162 HIV protein – host RNA interactions (FC > 4; Padj < 0,05) across 8 different HIV proteins.

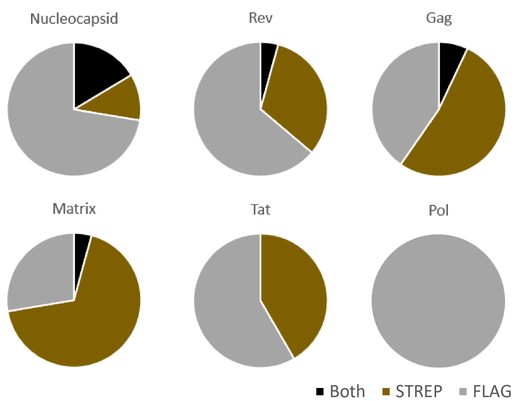


**Figure 2.** Pie diagram depicting the fraction of overlap between FLAG- and STREP-based RIPseq identified interactions.

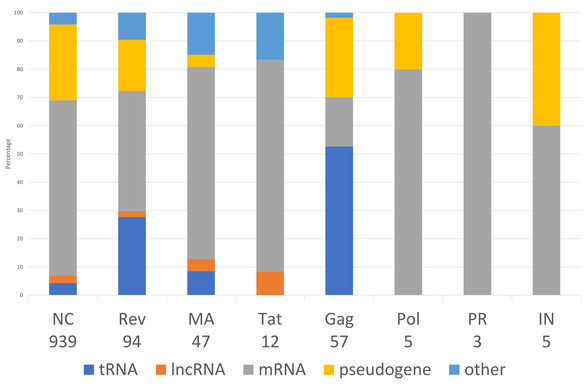


**Figure 3.** Biotype percentages of the identified host RNA interaction partners per HIV protein. HIV proteins with zero interactions are omitted. Numbers refer to the total interactions identified.

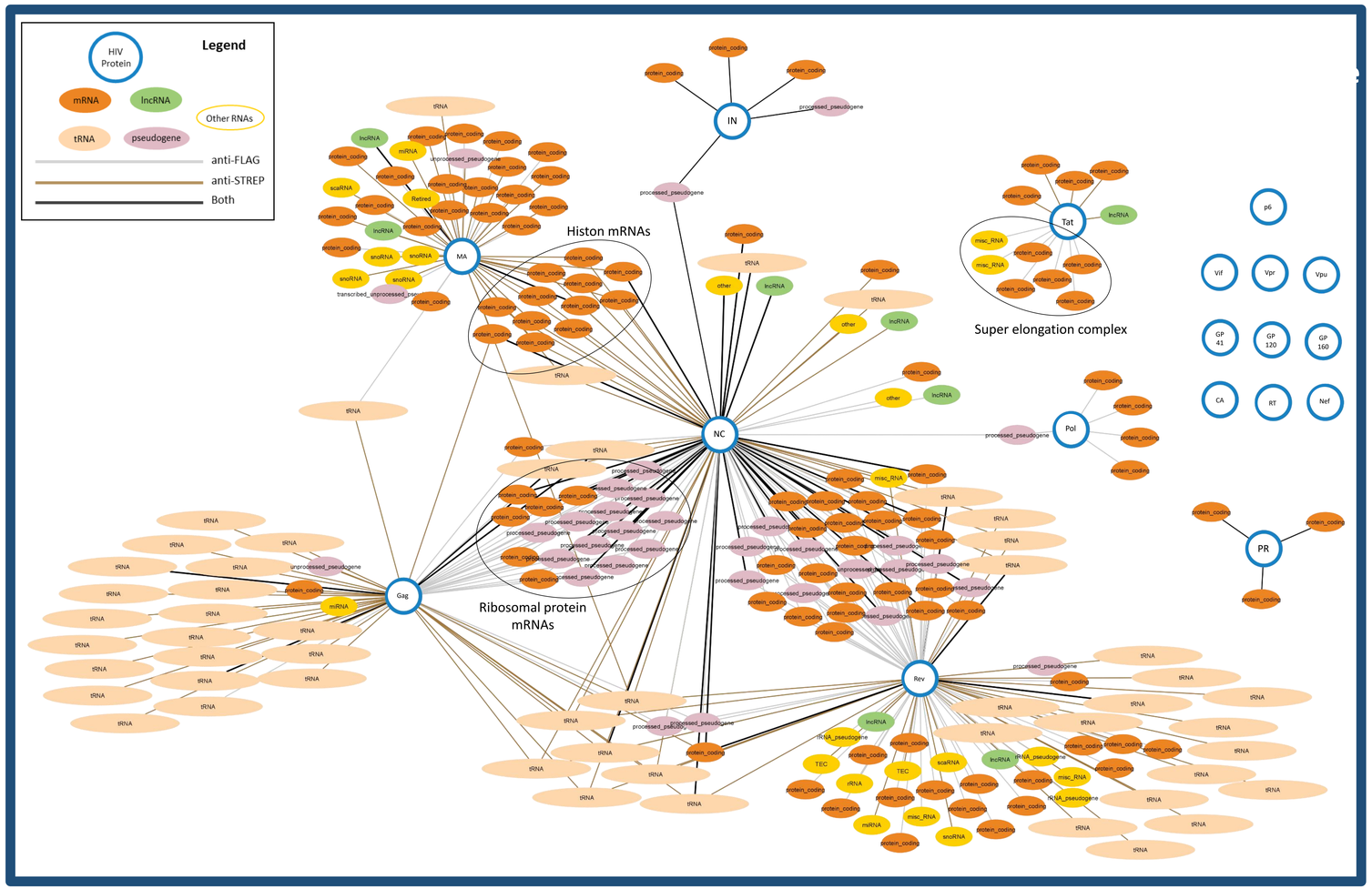


**Figure 4.** Cytoscape network including all interactions identified with either FLAG-based or STREP-based RIPseq.

## Interactome highlights

- Gag has a high preference for tRNA binding (53%); Rev (28%), Matrix (9%) and nucleocapsid (4%) also bind numerous tRNAs.
- Tat RNA interaction partners include mRNAs for which the translation protein products are known interactors of Tat within the super elongation complex (SEC).
- Matrix and nucleocapsid bind a cluster of histone mRNAs. Gag and Nucleocapsid share a cluster of ribosomal protein mRNAs.

## CONCLUSIONS

This unique and comprehensive dataset will broaden our understanding how HIV manipulates the RNA component of the host's cellular machinery during infection and can be mined to investigate potential new therapeutic strategies.

## ADDITIONAL INFORMATION

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