BLM Helicase Is Required for Efficient HIV Infection in Macrophages

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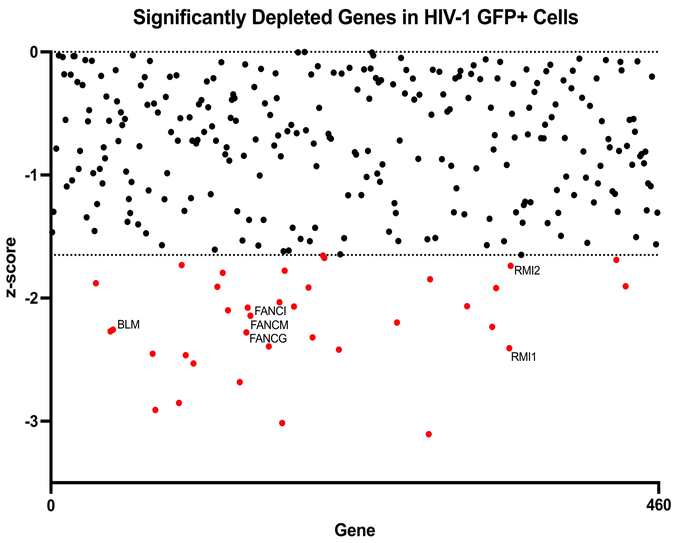
BACKGROUND

During infection HIV-1 engages host DNA damage responses (DDRs) at multiples steps of the viral replication cycle. Many DDR factors interact with HIV-1 in early infection leading up to the establishment of integrated provirus. Exactly how DDRs are modulated and the extent to which they negatively or positively influence infection remain unclear.

This project aims to identify novel host DDR/HIV-1 interactors and explore mechanisms by which the facilitate or restrict viral integration.

METHODS

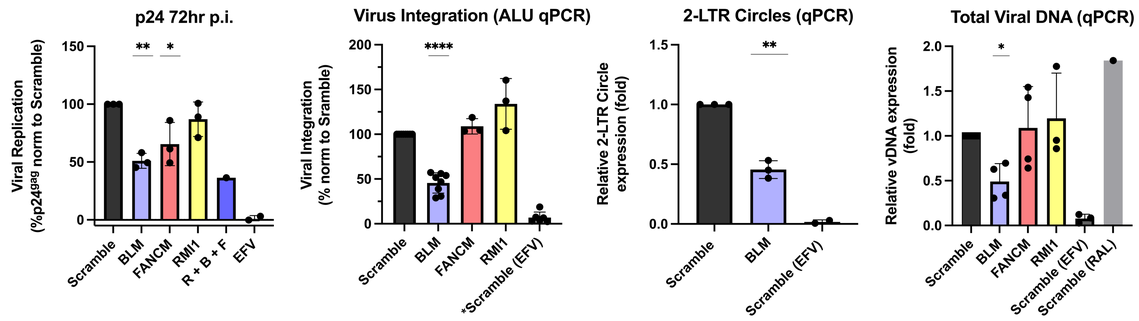
- Using a DDR-specific CRISPR/Cas9 screen, we identify DDR factors that influence HIV-1 infection in CHME cells
- Validation of screen hits was performed in primary monocyte-derived macrophages
- Knockdowns were done using a double-dose of pooled siRNAs (Horizon) and confirmed via qPCR and WB



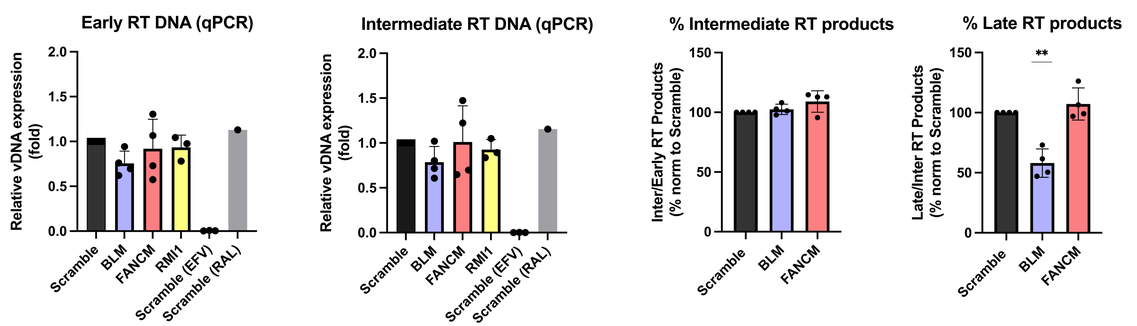
- ❖ Knockdown of BLM helicase in primary macrophages inhibits HIV-1 late reverse transcription and integration
- ❖ BLM binds HIV-1 DNA during reverse transcription, before integration

RESULTS

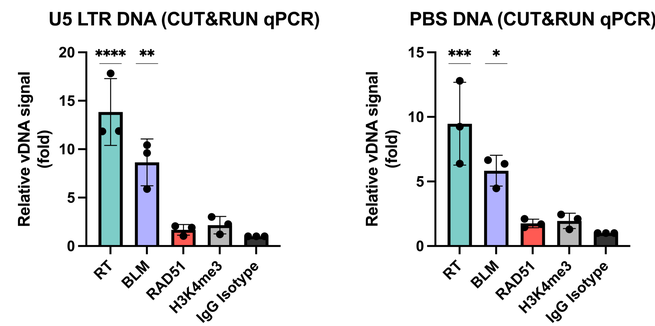
- Knockdown of BLM in MDMs results in 50% reduction of viral replication (p24gag), viral integration (ALU-qPCR), episomal vDNA (2-LTR circles), and total viral DNA (late RT products)



- Knockdown of BLM in MDMs did not affect early or intermediate reverse transcription products



- BLM binds HIV-1 DNA near U5 LTR during reverse transcription in CHME cells



CONCLUSIONS

- BLM helicase facilitates completion of reverse transcription and integration in macrophages
- BLM binds HIV-1 DNA intermediates concomitant with the 2nd strand transfer during reverse transcription
- Future studies will assess mechanisms of binding and timing during RTC to PIC transformation, as well cell type differences of DDR engagements between CD4+ T cells and macrophages

Funding
- **NIH T32 AI007309**
- **NIH R01 AI187175**
- **NIH R01 DA055488**
- **CFAR P30 AI042853**

PLAIN LANGUAGE SUMMARY

We discover a new interaction between HIV-1 and an essential host DNA damage response protein, BLM, that appears to facilitate efficient infection in cells that are capable of harboring persistent latent reservoirs.