Pan-retroviral nucleocapsid-mediated phase separation regulates genomic RNA positioning and trafficking

Article

Authors

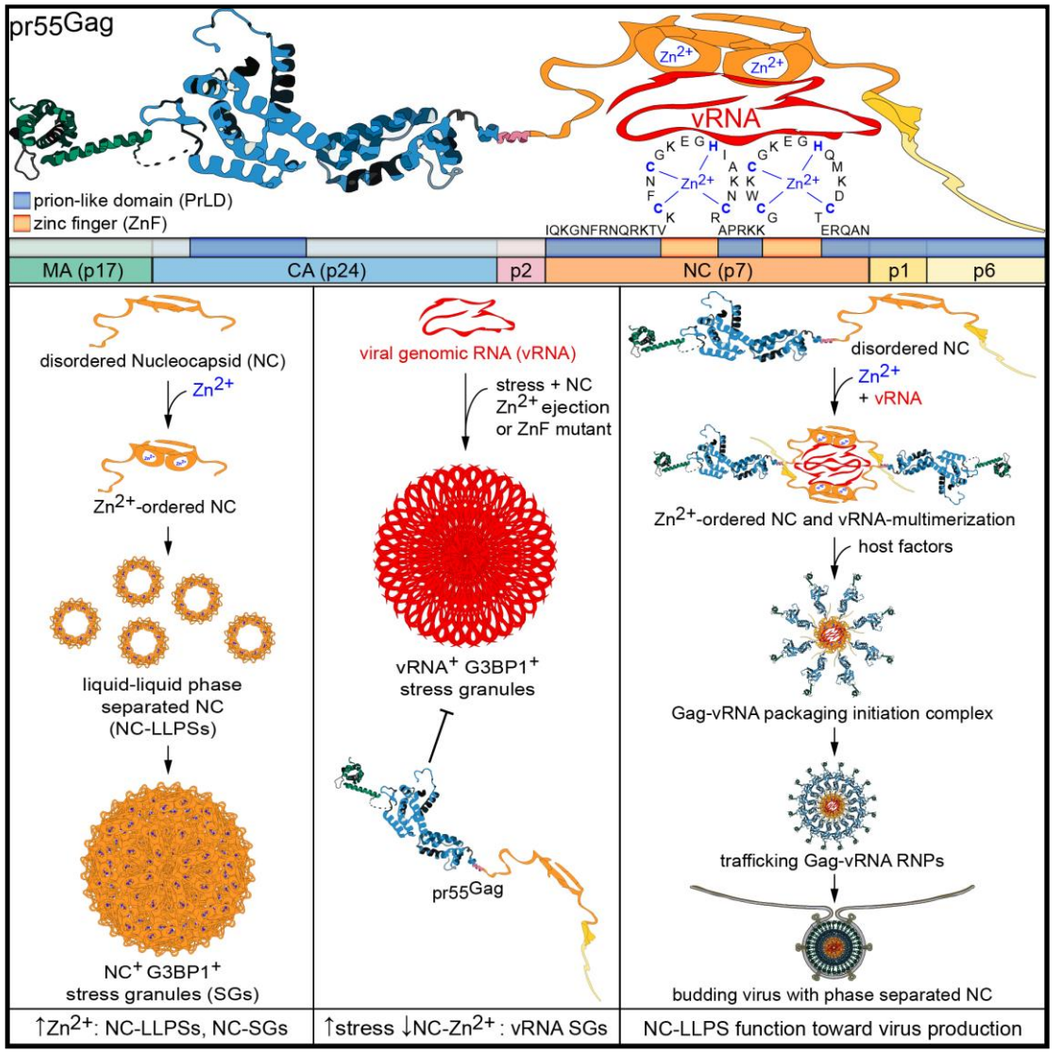
Anne Monette, Meijuan Niu, Lois Chen, Shringar Rao, Robert James Gorelick, Andrew John Mouland

Correspondence

anne.monette@mail.mcgill.ca (A.M.), andrew.mouland@mcgill.ca (A.J.M.)

In Brief

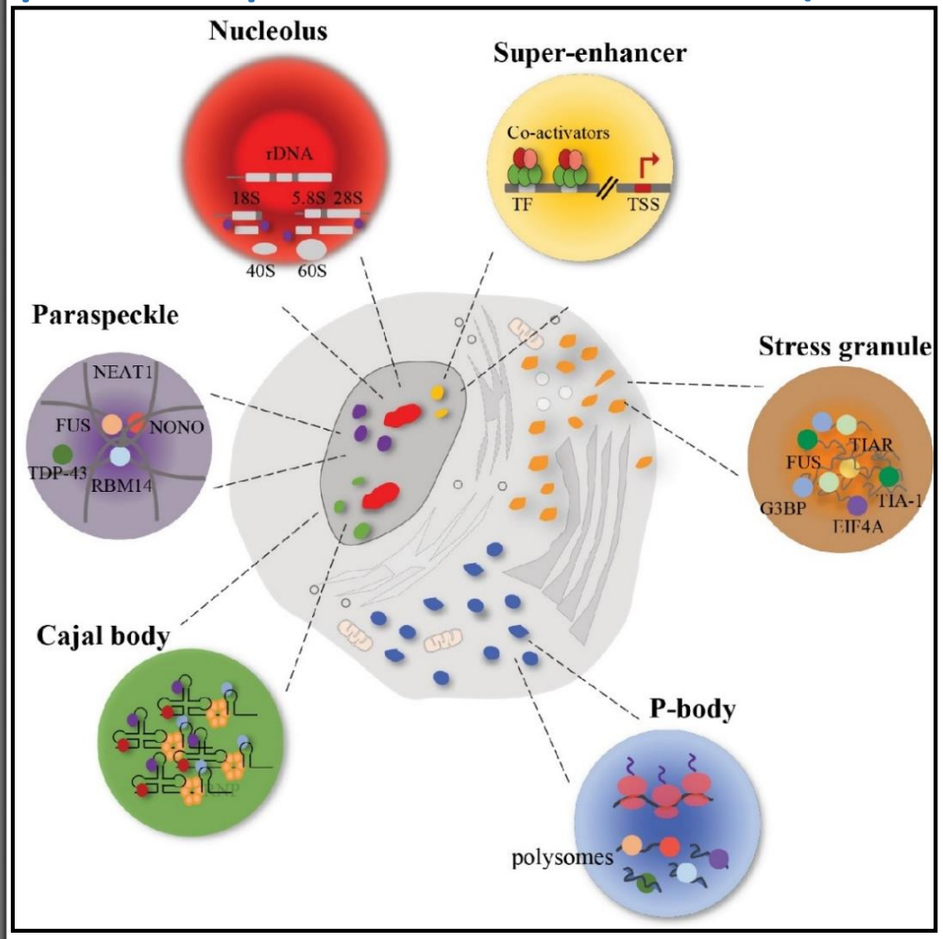
Monette et al. discover a high degree of conservation of zinc-finger embedded, intrinsically disordered prion-like domains across retrovirus Gag proteins. These domains within the Gag Nucleocapsid regulate the formation of zinc-dependent liquid-liquid phase condensates and stress granules in HIV-1-expressing cells to induce repositioning of the viral genomic RNA.

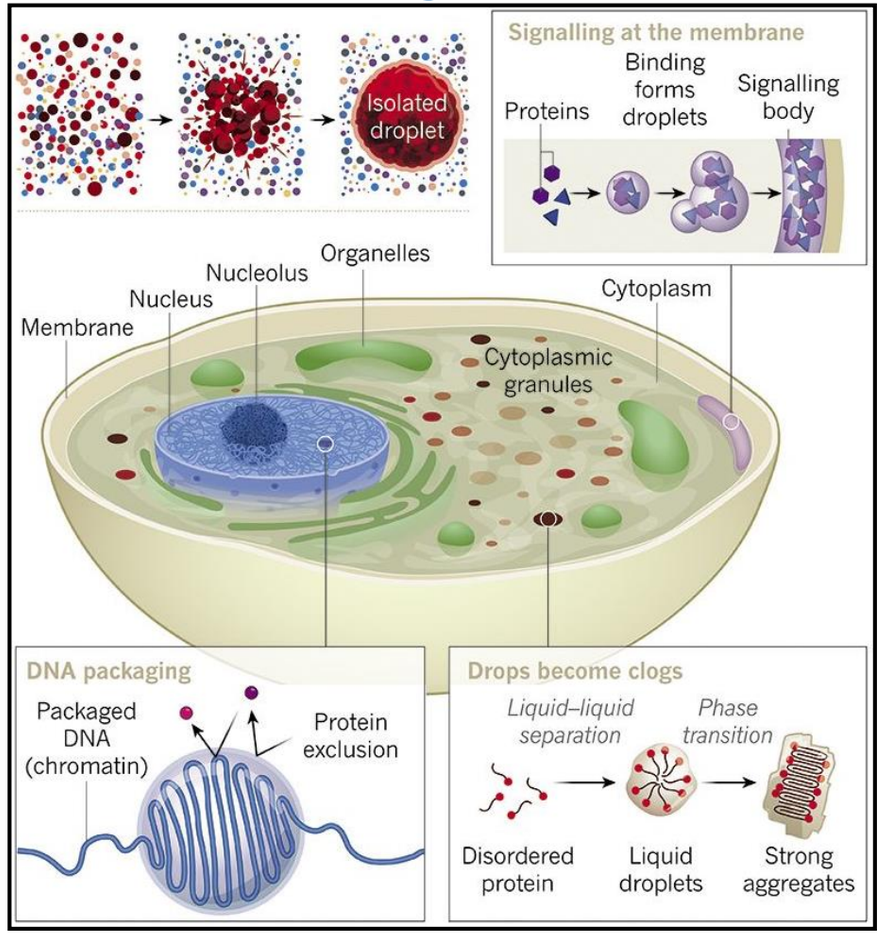


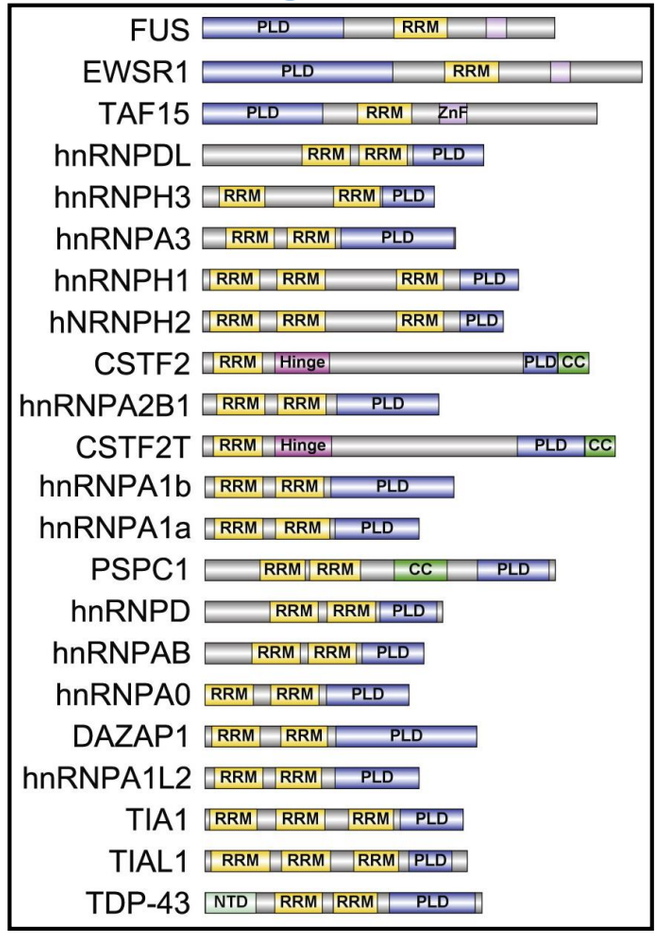
Highlights

- Retroviral Gag proteins have conserved intrinsically disordered prion-like domains
- Pan-retrovirus family nucleocapsid proteins induce liquid-liquid phase separation
- Nucleocapsid protein phase separation and stress granule assembly is Zn^{2+} dependent
- Zn^{2+} chelators and ejectors induce nuclear repositioning of the genomic RNA

Membraneless organelles are formed by liquid-liquid phase separation of disordered protein prion-like domains (PrLDs), RNA-binding motifs (RRMs) & Zinc fingers (ZnFs)



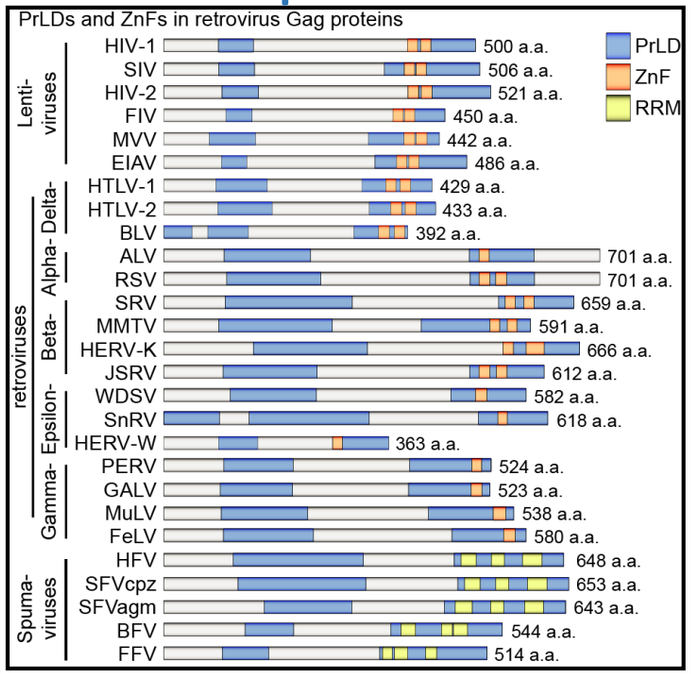


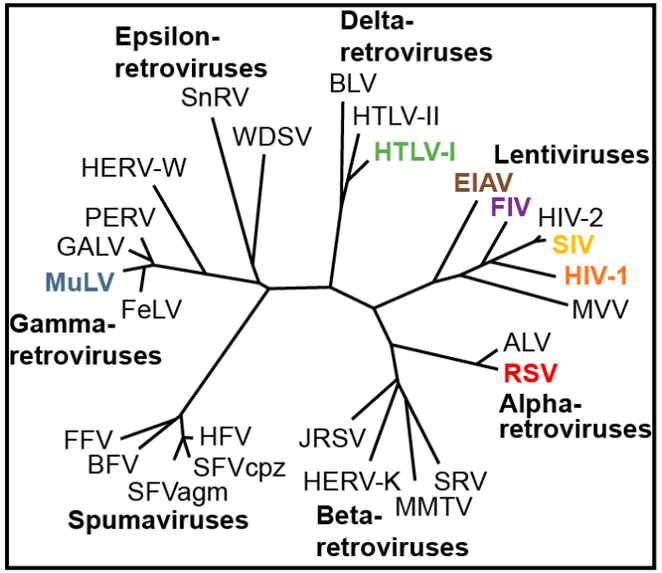


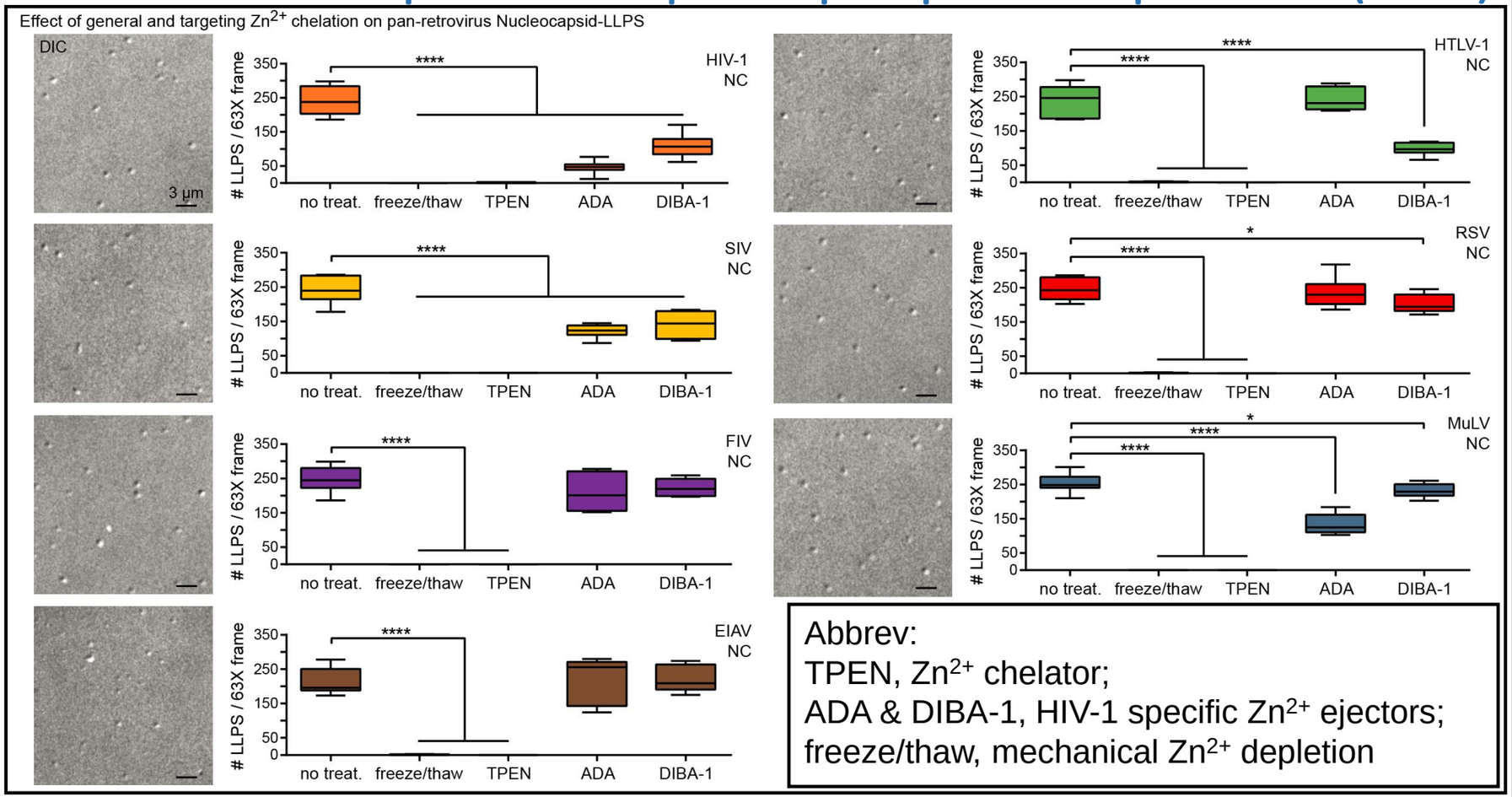
Like HIV-1 Nucleocapsid (NC), cellular RNA-binding, helicases and chaperones, such as G3BP1, TIAR, TIA-1, DDX6, TDP-43, FUS/TLS, Tau, and hnRNP proteins, are also components of stress granules (SGs) and P-bodies (PBs), can be Zn^{2+}-regulated, and form liquid-liquid phase separation (LLPS) condensates due to their low-complexity, intrinsically disordered prion-like domains (PrLDs).

Elie Dolgin. Nature. 2018 Mar 15;555(7696):300-302.
Verdile V. et al. Front Genet. 2019 Mar 22;10:173.
Wang J. et al. Cell. 2018 Jul 26;174(3):688-699.
Rao S. et al. RNA. 2018 Feb;24(2):219-236.
Kedersha N. et al. J Cell Biol. 2016 Mar 28;212(7):845-60.
Garnier C. et al. Sci Rep. 2017 Jul 28;7(1):6812.
Rayman JB. et al. Cell Rep. 2018 Jan 2;22(1):59-71.
Panas MD. et al. J Cell Biol. 2016 Nov 7;215(3):313-323.

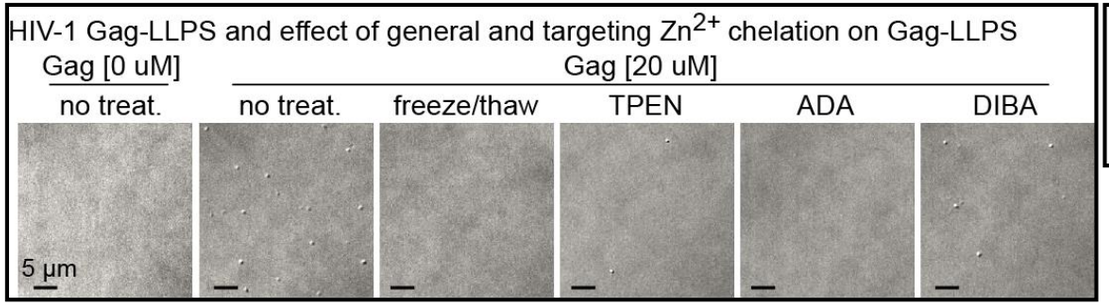
Pan-retrovirus Gag proteins have conserved PrLDs, ZnFs, and RRMs, and undergo Nucleocapsid-mediated ZnF and Zn^{2+}-dependent liquid-liquid phase separation (LLPS)







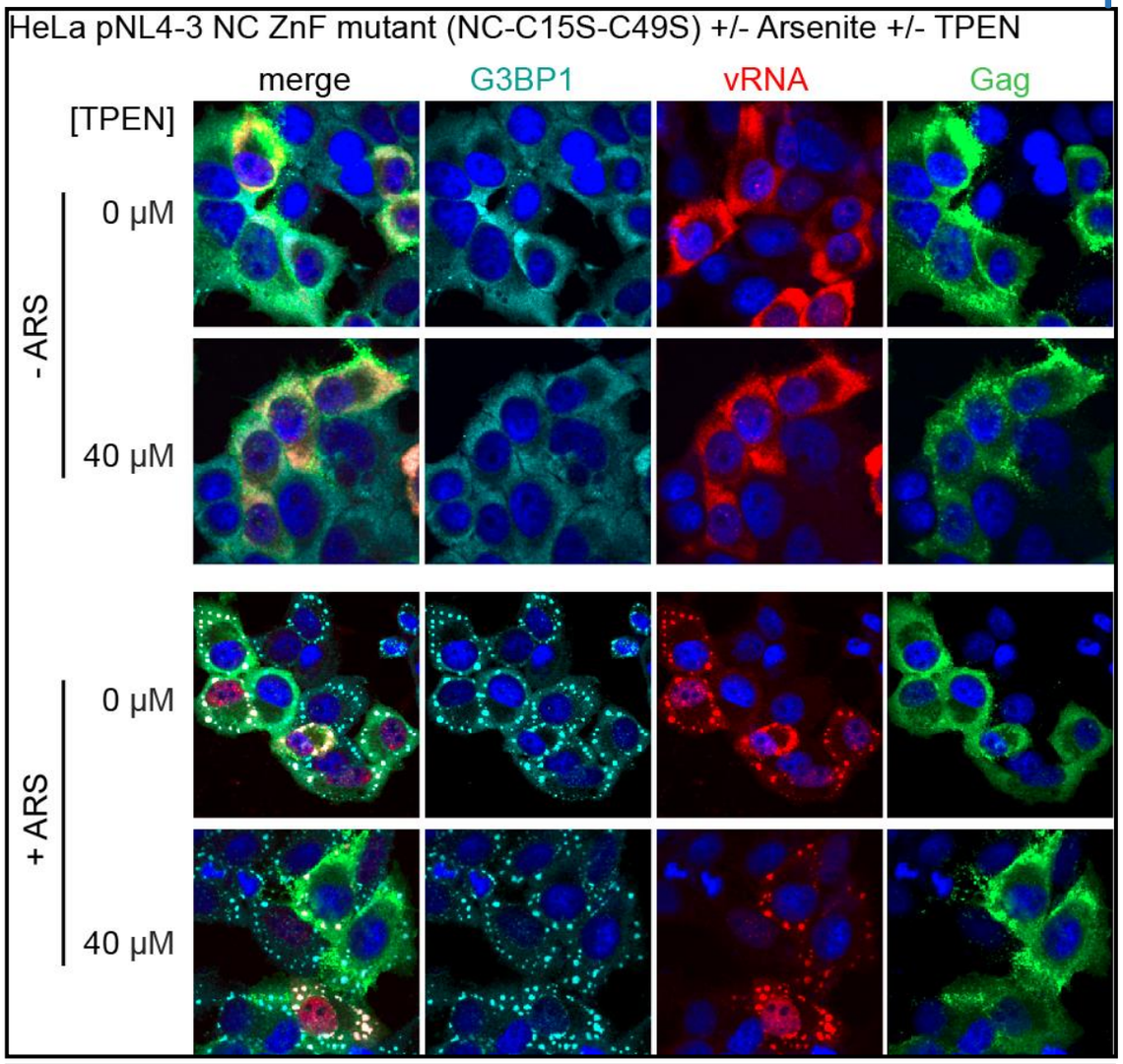
Abbrev:
TPEN, Zn^{2+} chelator;
ADA & DIBA-1, HIV-1 specific Zn^{2+} ejectors;
freeze/thaw, mechanical Zn^{2+} depletion

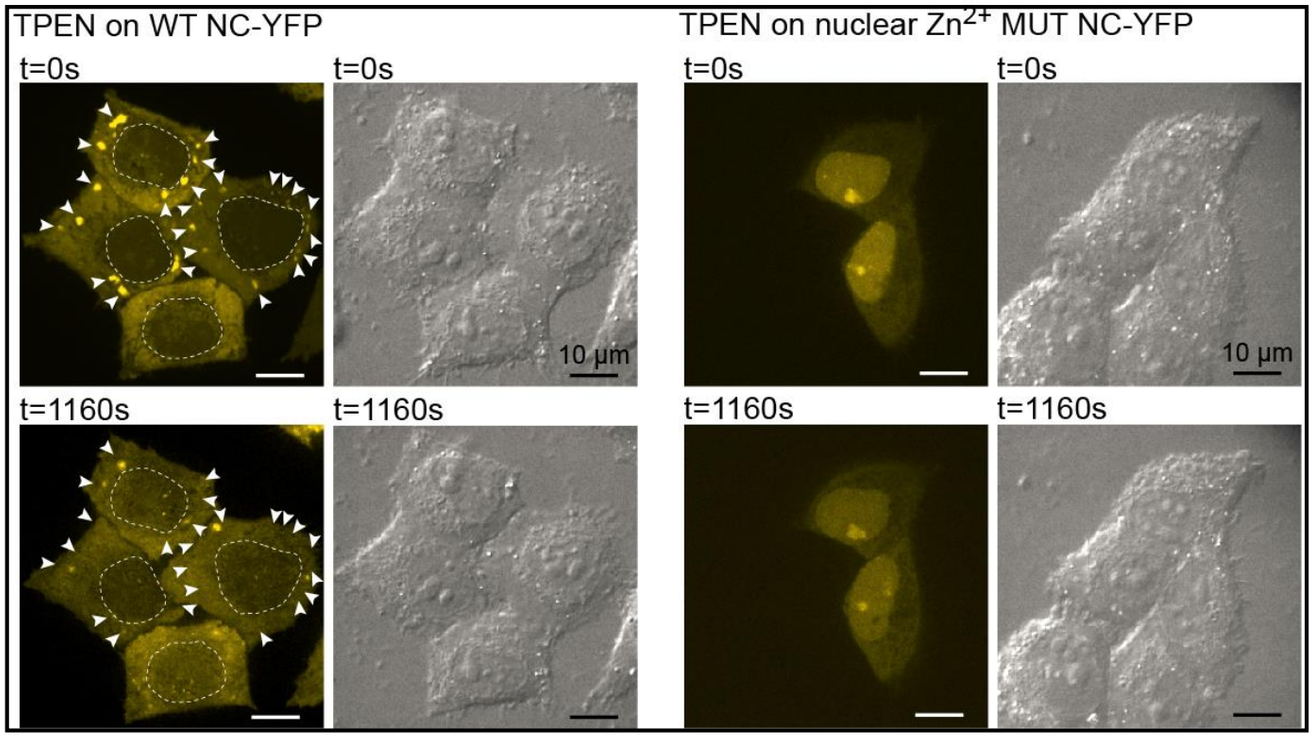


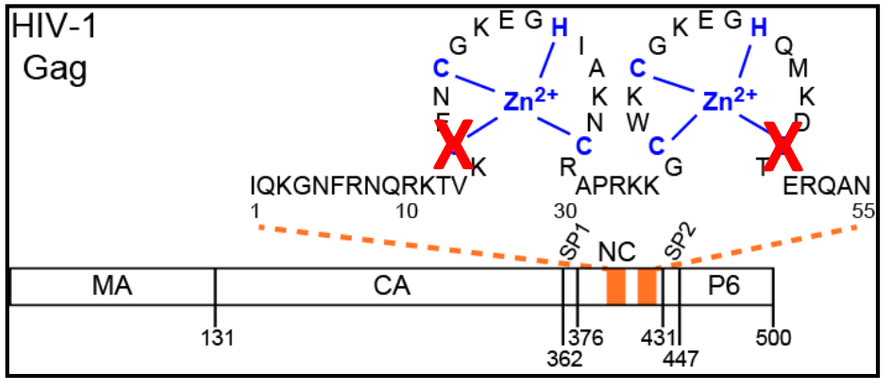
Abbrev: PrLD, prion-like domain;
RRM, RNA-binding motif;
ZnF, zinc finger

Linial ML. et al. J Virol. 1999 Mar;73(3):1747-55.
Weiss RA. et al. Retrovirology. 2006 Oct 3;3:67.
Monette A. et al. Cell Rep. 2020 Apr 21.

NC ZnF mutants cause SG-vRNA accumulation & reverse HIV-1 blockade on SGs & NC-LLPS formed in live cells are ZnF-dependent and Zn^{2+}-chelation sensitive







Abbrev: Zn^{2+}, zinc; ZnF, zinc finger; SG, stress granule; vRNA, viral genomic RNA; ARS, arsenite; LLPS, liquid-liquid phase separation

Valiente-Echeverría F. et al. Nat Commun. 2014 Sep 17;5:4819.
Cinti A. et al. mBio. 2016 Mar 29;7(2):e00329.
Monette A. et al. Cell Rep. 2020 Apr 21.

Acknowledgements

Andrew Mouland
Meijuan Niu
Lois Chen
Shringar Rao
Sergio Alpuche
Paola Guizar
Gabriel Guajardo Contreras
Ana Luiza Abdalla

Robert Gorelick
Donald Johnson
Cathy Hixson

Imed Gallouzi
Maureen Oliviera
Ilinca Ibanescu
Cesar Collazos
Bonnie Spira
Colin Crist
Solene Jamet
Henry Yu
Stephane Richard
Rongtuan Lin
Chia-Hao Chang
Christian Young
LDI cell imaging facility

CIHR IRSC
Canadian Institutes of Health Research
Instituts de recherche en santé du Canada

NIH
National Institutes of Health

During the COVID-19 pandemic, virus transmission is now often described in terms of “aerosols” or “droplets”. Bioinformatics, *in vitro*, and cellular immunofluorescence and biochemical approaches are used to explore pan-retrovirus assembly in terms of liquid-liquid phase separation. Photography by Anne Monette.