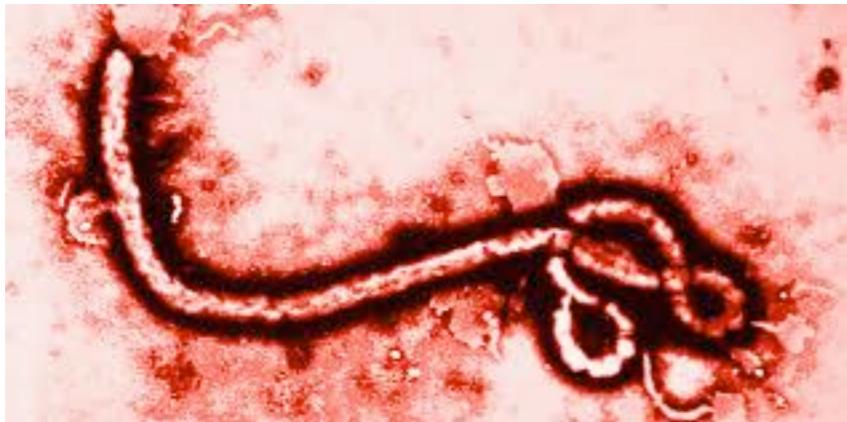
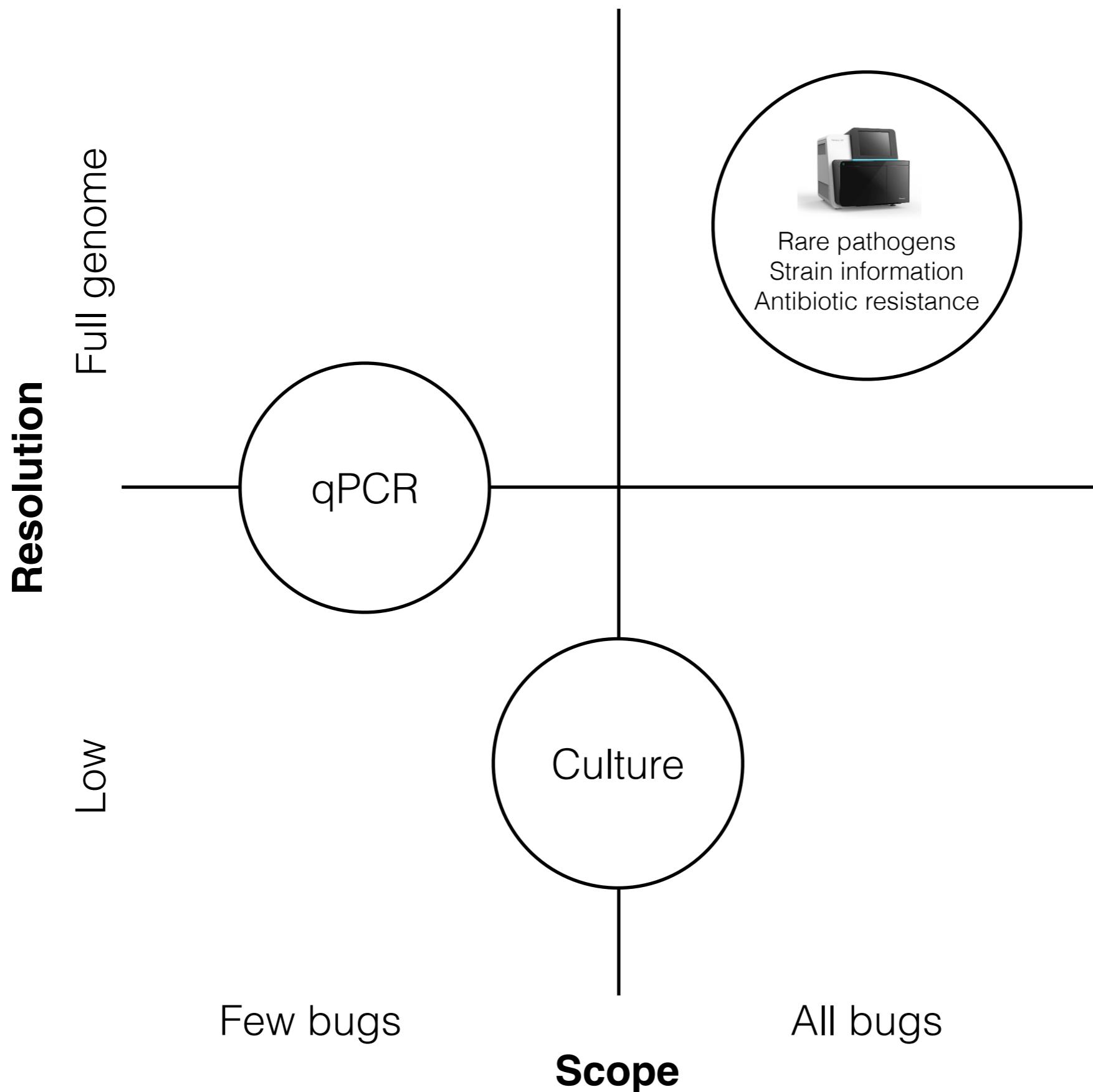


Monitoring the human infectome

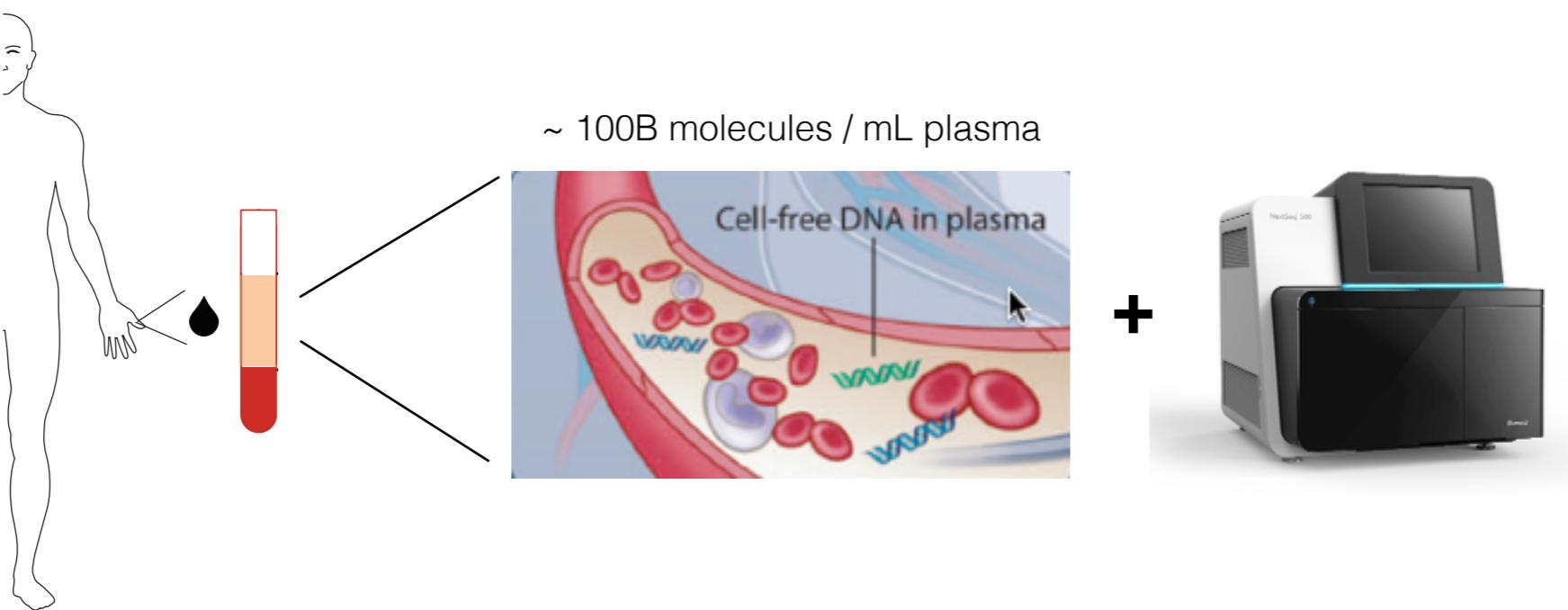
Lance Martin
Howard Chang & Steve Quake Labs
EpiBio 12/12/14



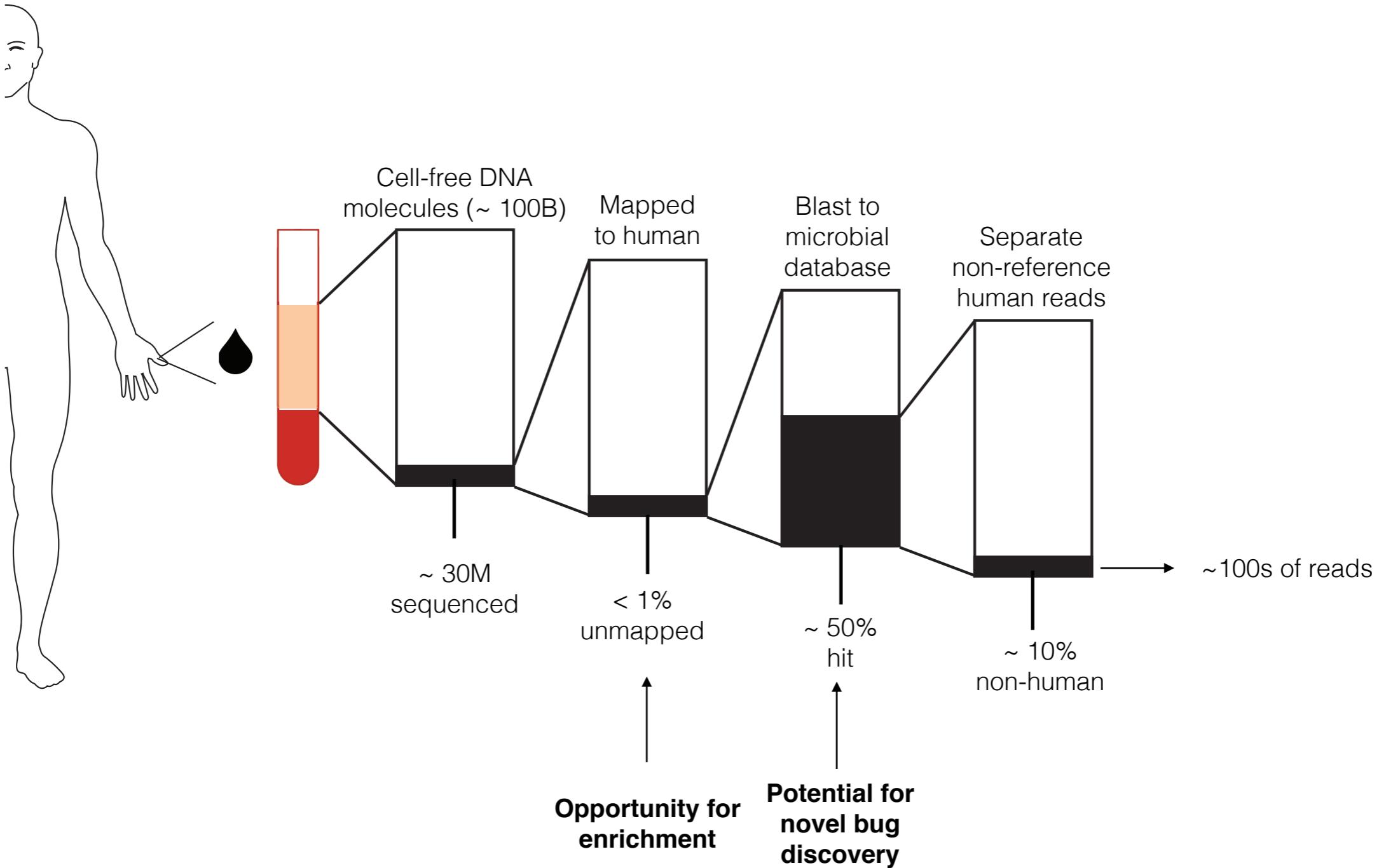
Non-invasive detection of any pathogen?



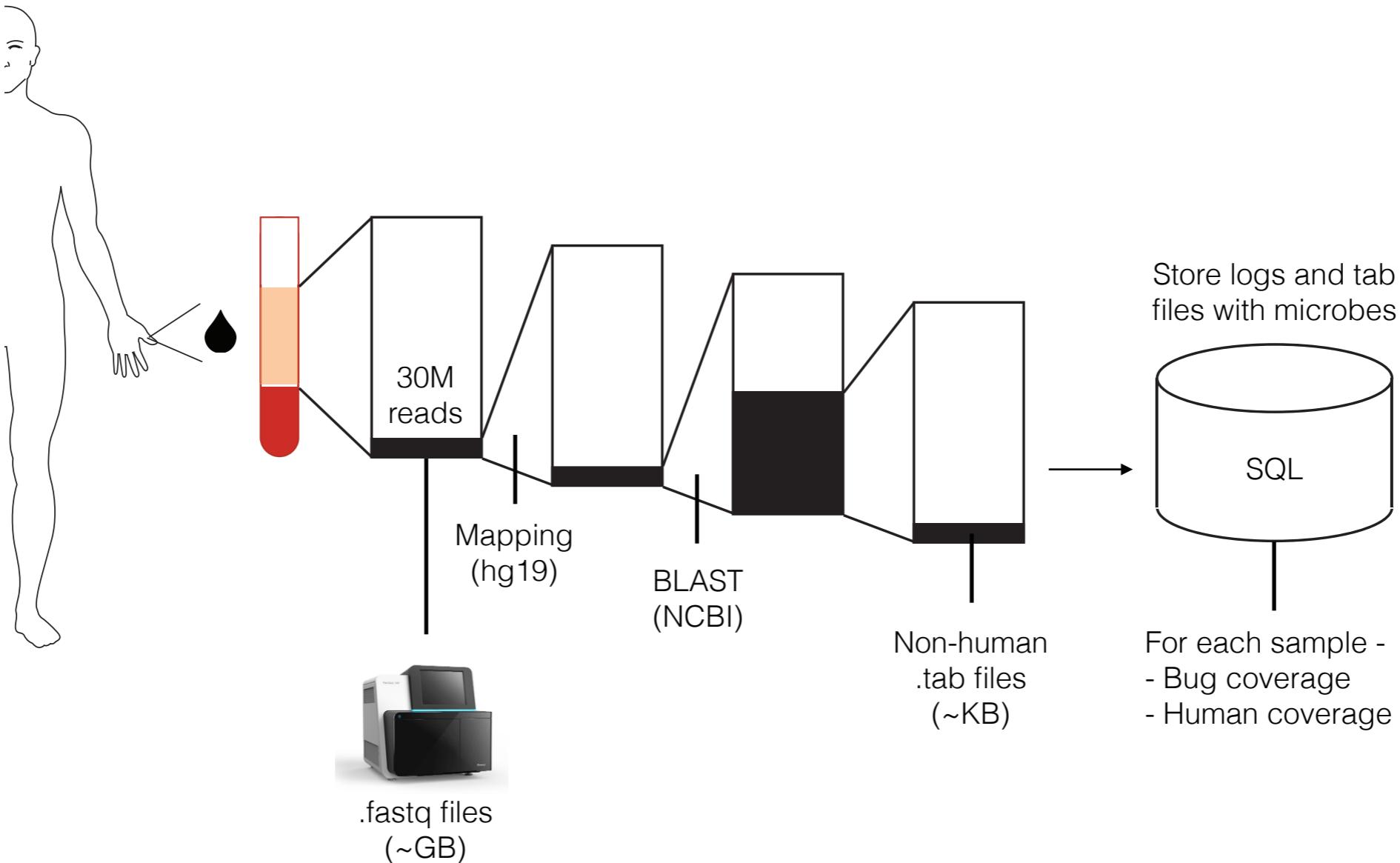
Consider cell-free DNA for non-invasive monitoring.



Human microbiome in cell-free DNA.

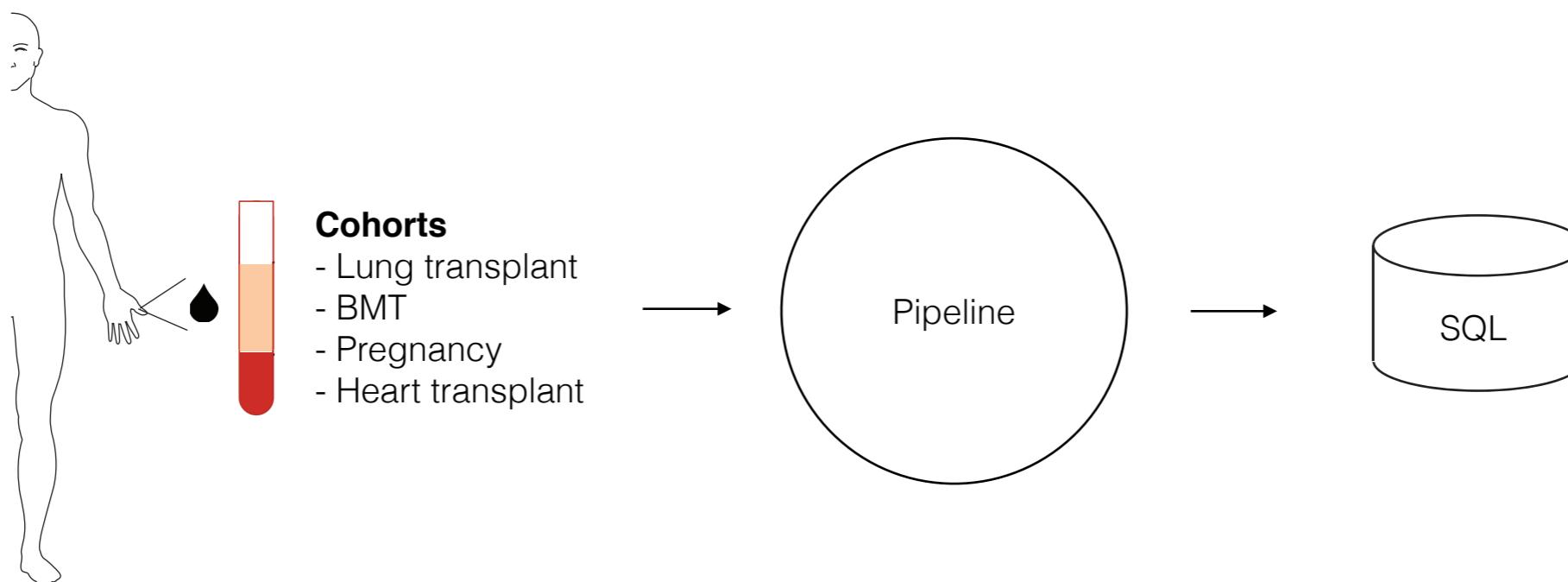


Pipeline for isolating microbial reads in cell-free data.



Mine existing datasets!

1000s of samples with cfDNA collected



Browser for visualization and navigation.

Cohorts

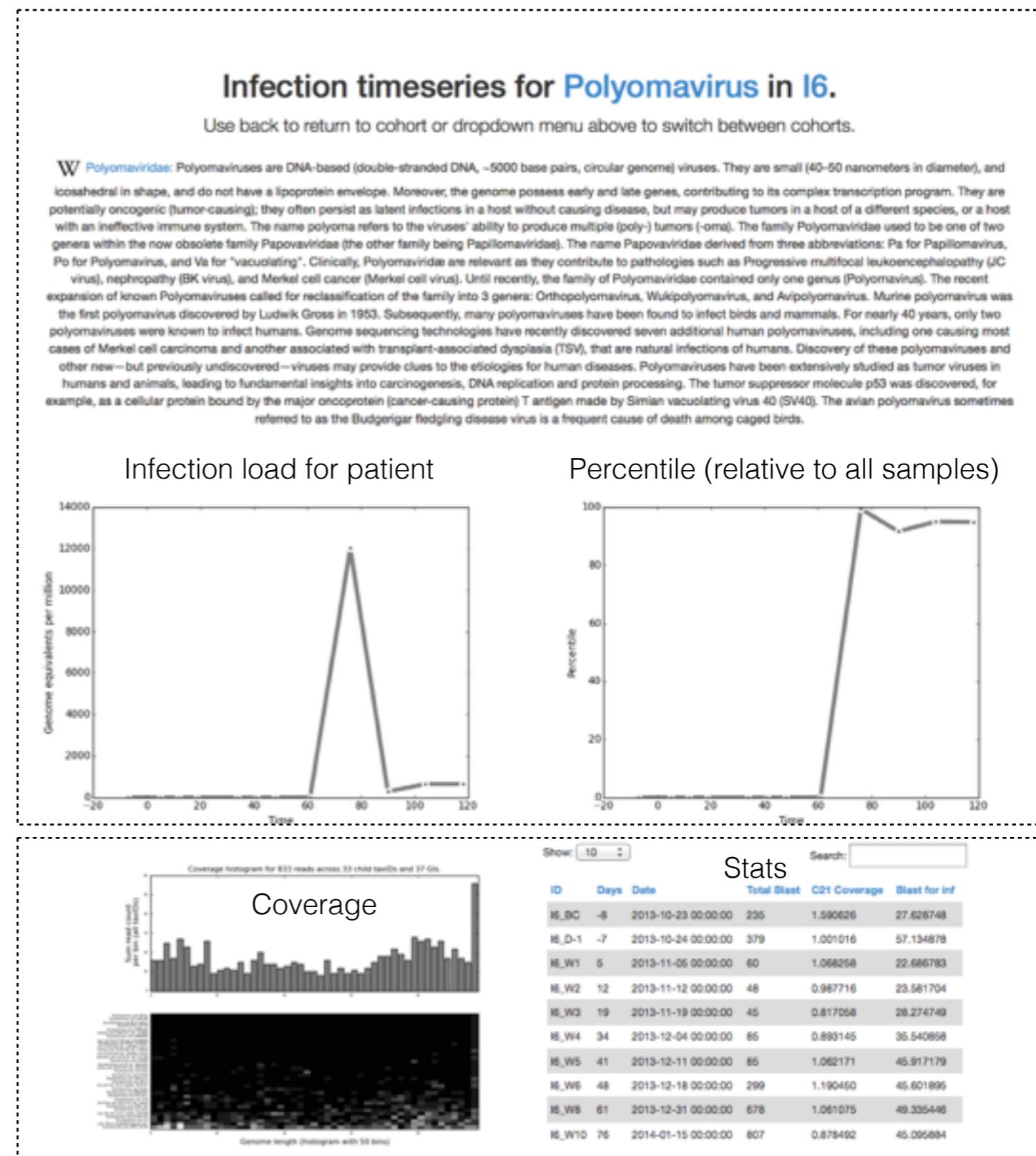
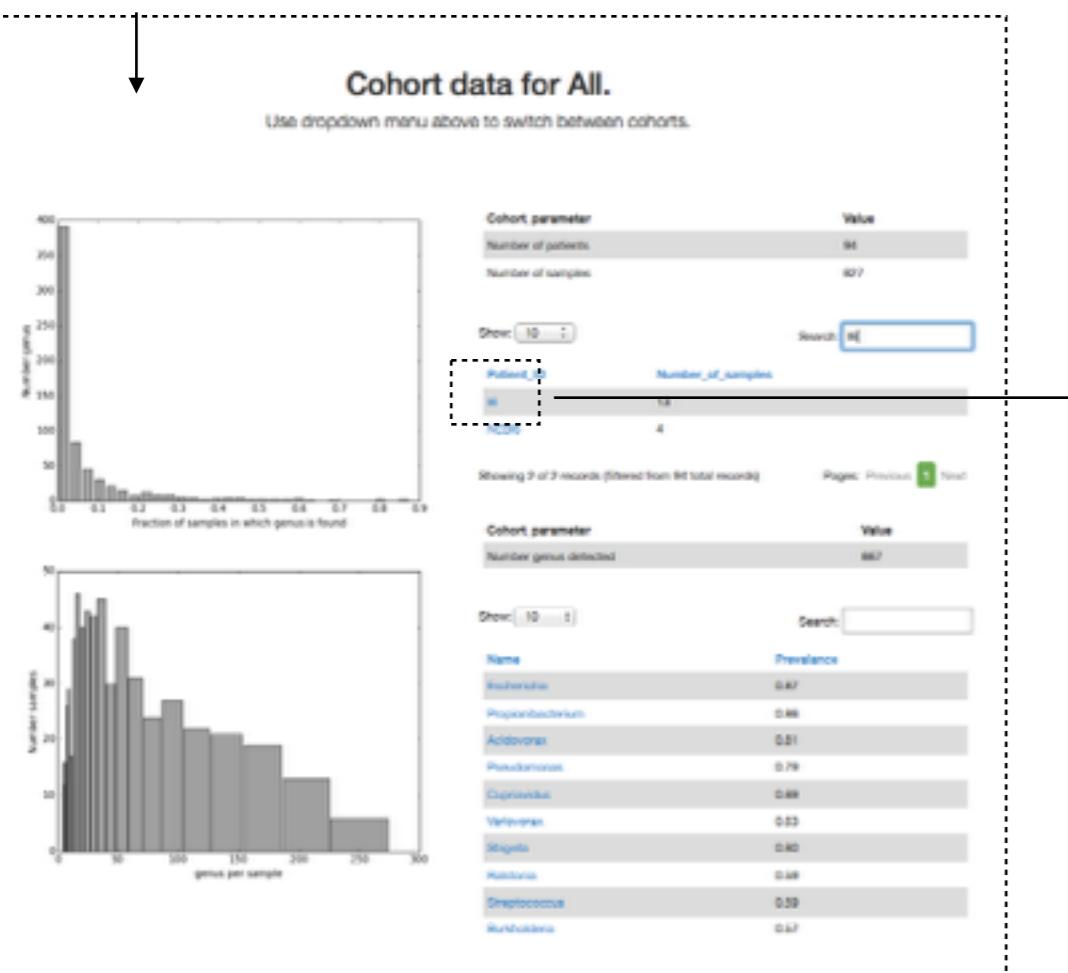
- Lung
- BMT
- Pregnancy
- Heart

Infectome Explorer Cohorts - Taxonomic level -

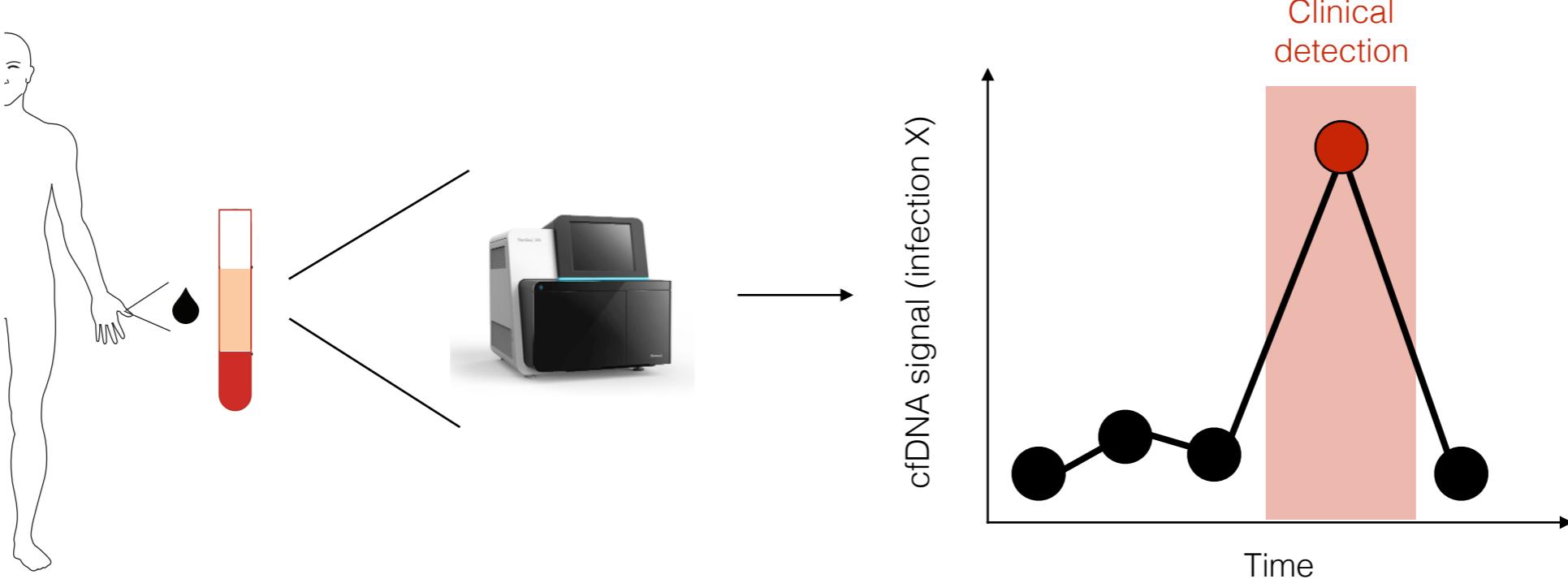
Welcome to the Infectome Explorer.

Choose a cohort:

All cohorts BMT Lung Pregnancy Ebola Stanford clinic Biopsy Pregnancy RNA CFS

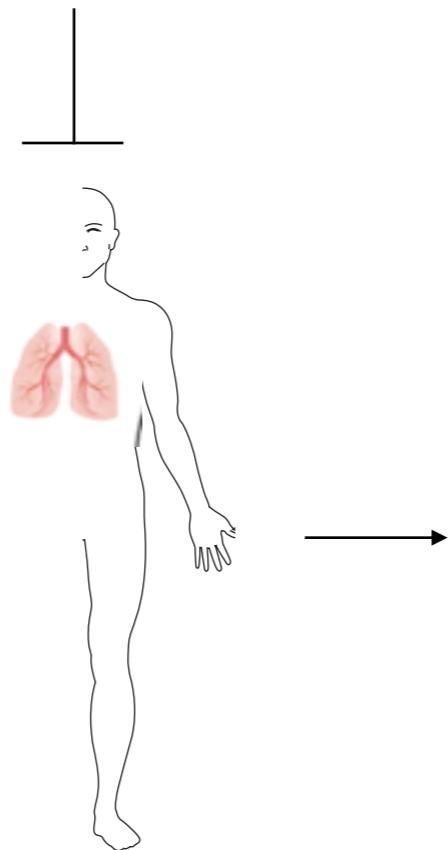


But how to know if it's useful?



Extensive infection testing in transplant cohorts.

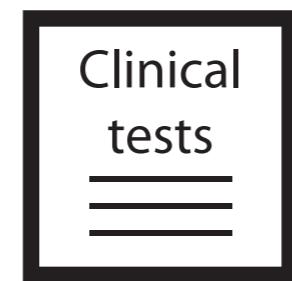
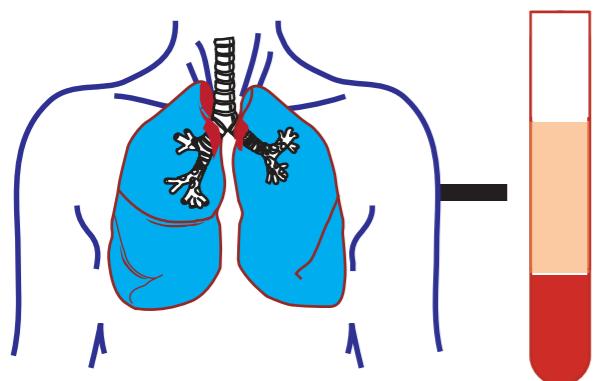
Immunosuppression



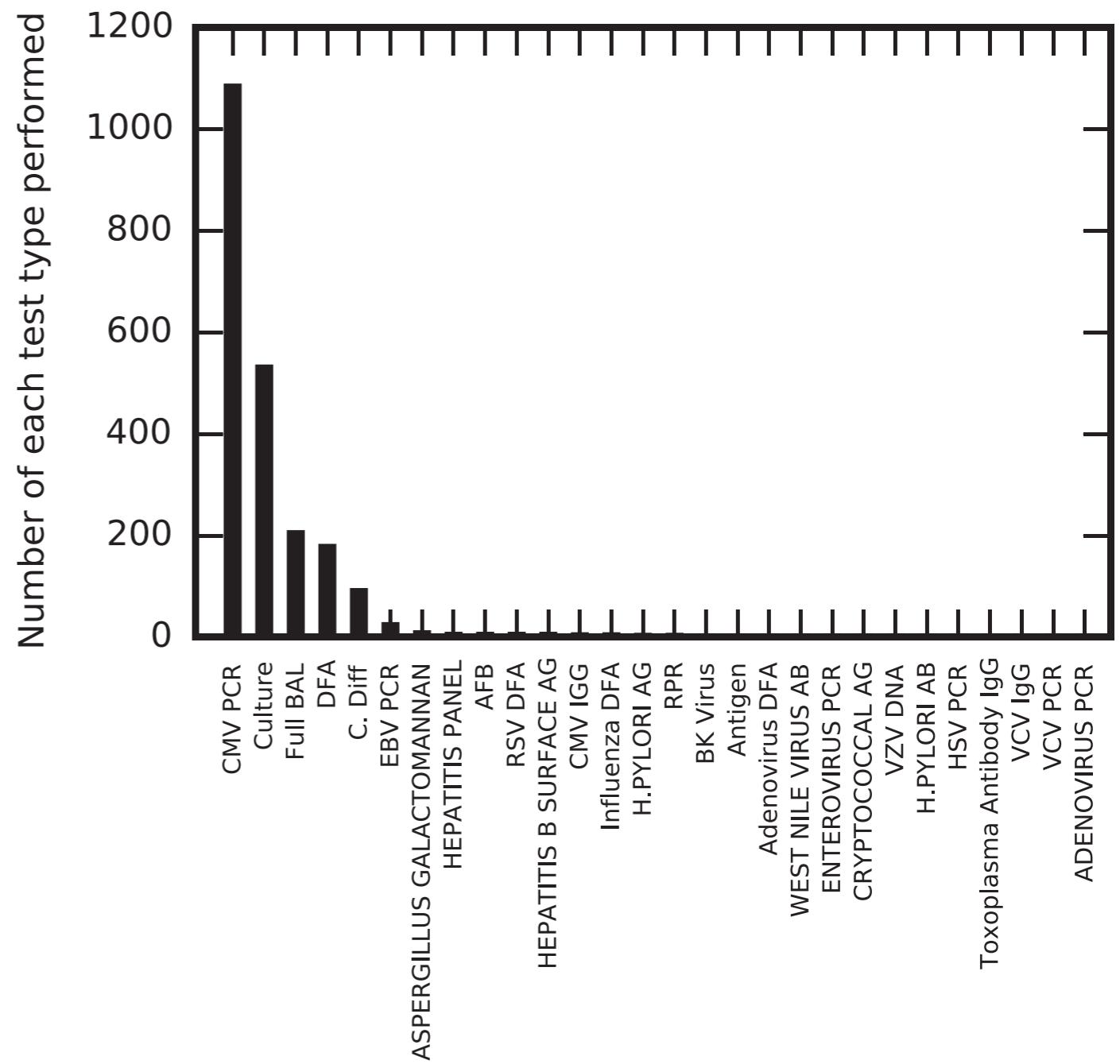
- (1) Susceptibility to exogenous infections
 - Hospital acquired
- (2) Resurrection of latent viruses
 - Herpesviruses
- (3) Infection (e.g., viruses) with graft
 - CMV (lung)

Look back at infection clinical history of our cohorts.

Lung Transplant (431 samples collected and processed) -

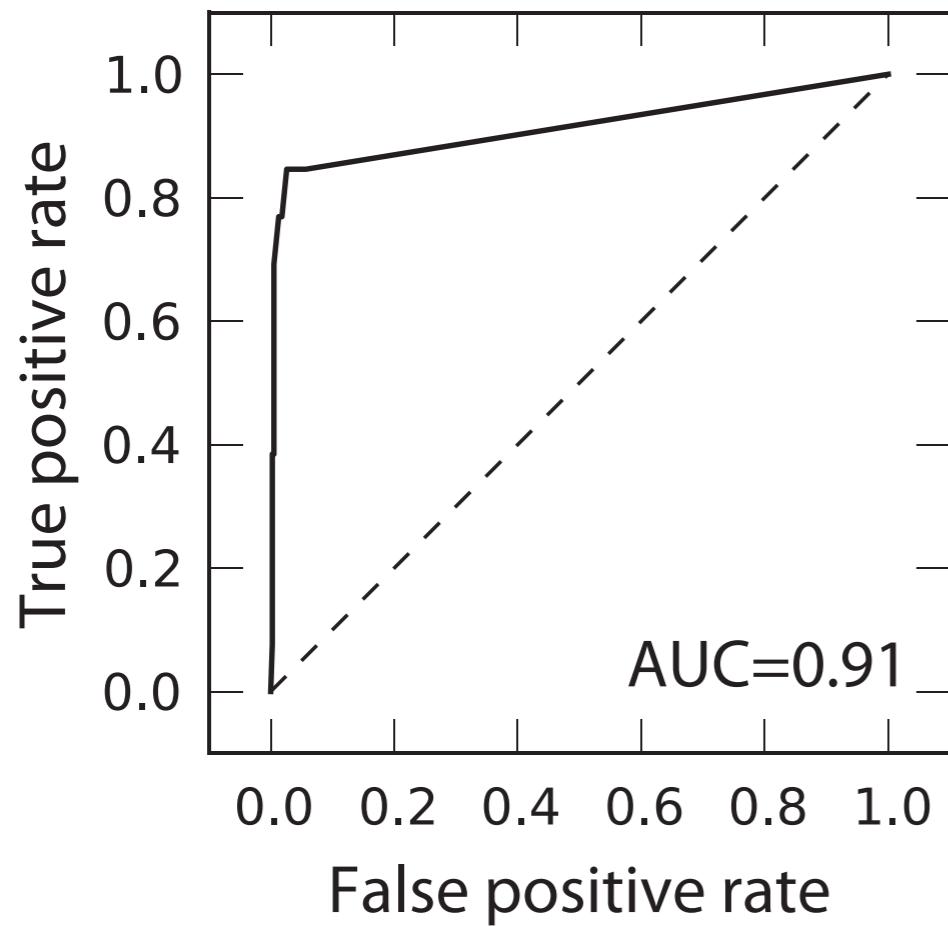


Thousands of tests recorded (~35k bug measurements) -

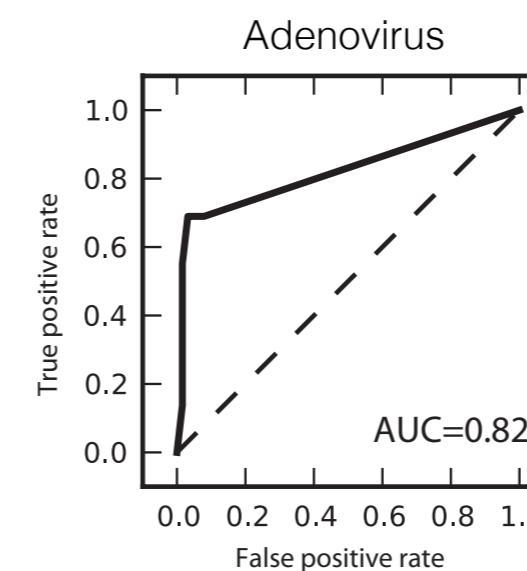


Performance on viruses.

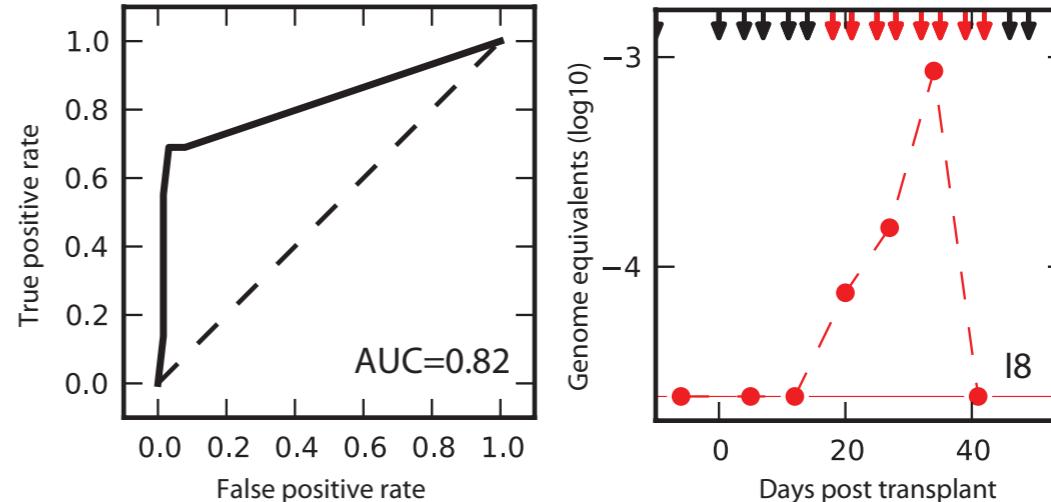
CMV in lung cohort -



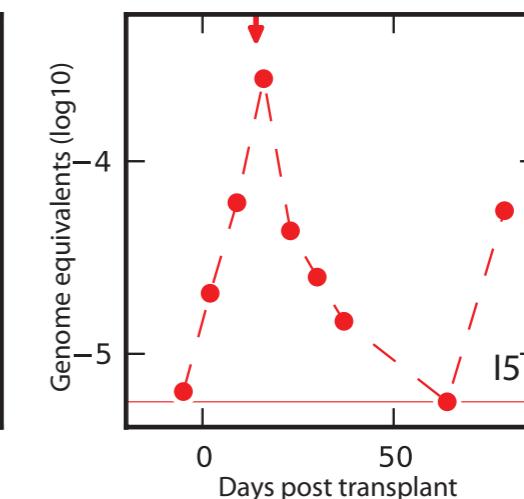
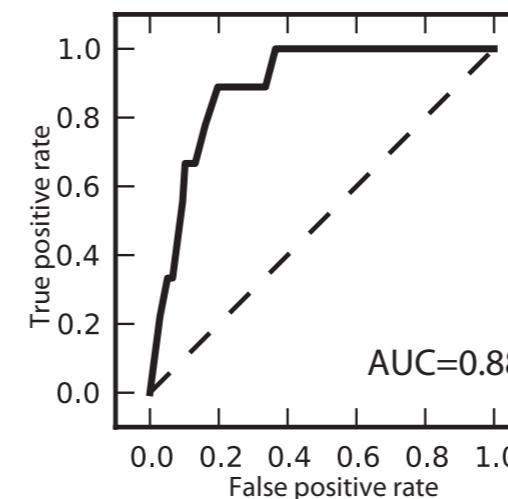
Bone marrow cohort -



Example patient with
clinical tests (pos, neg)

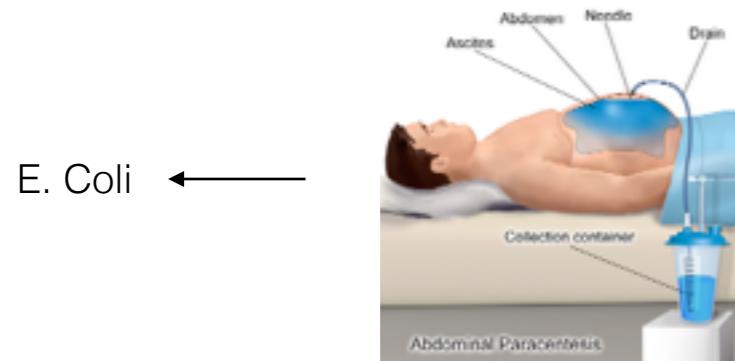


CMV

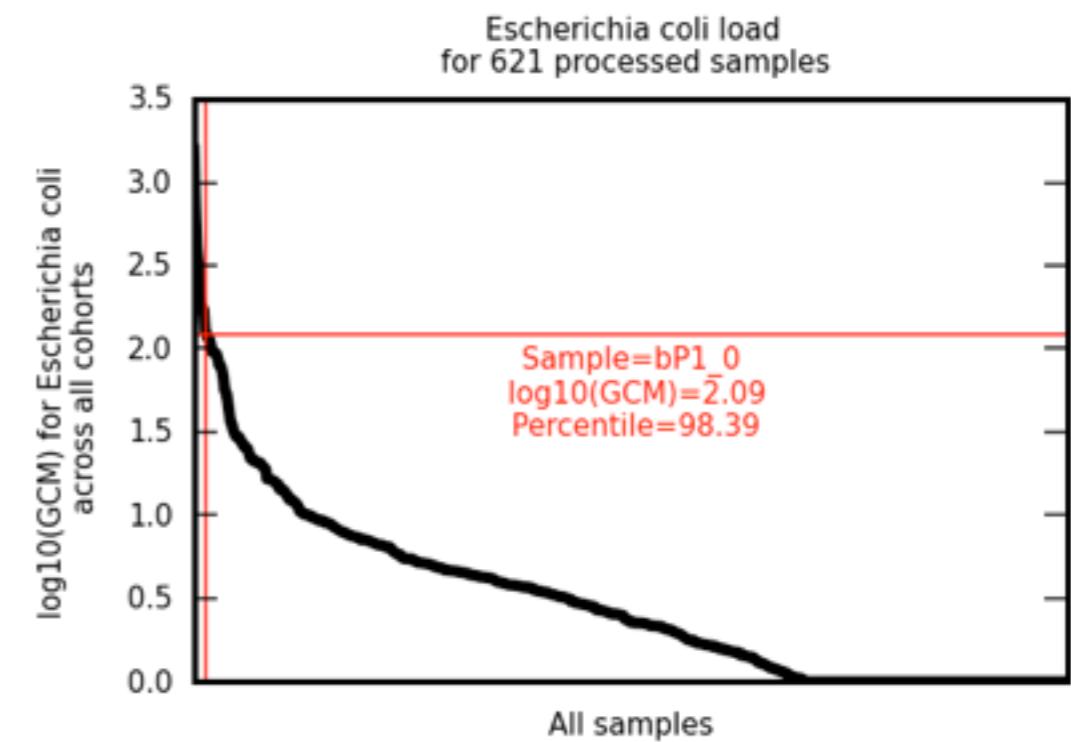
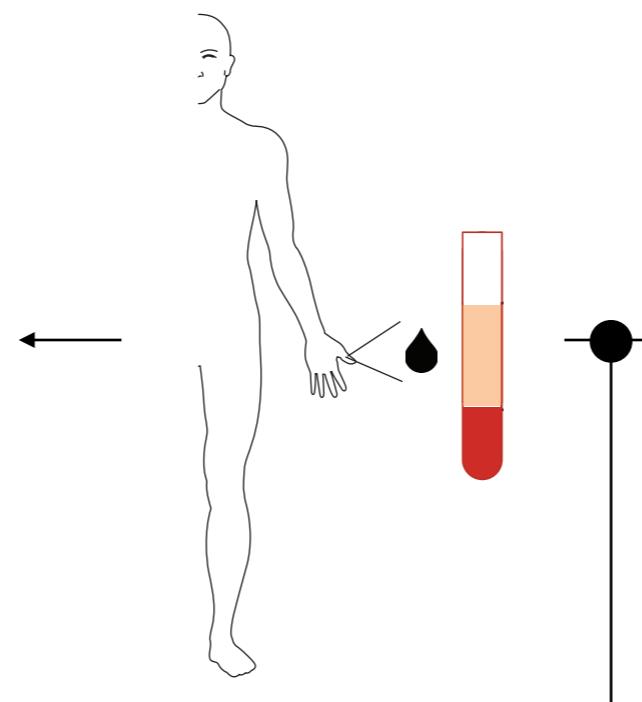


Performance on microbes (detected in deep tissues).

Stanford Pathology -
Biopsy and MALDI-tof



E. Coli



Ranking of all bugs detected in the sample -

Sorted infection data for bP1_0.

Use back to return to cohort or dropdown menu above to switch between cohorts.

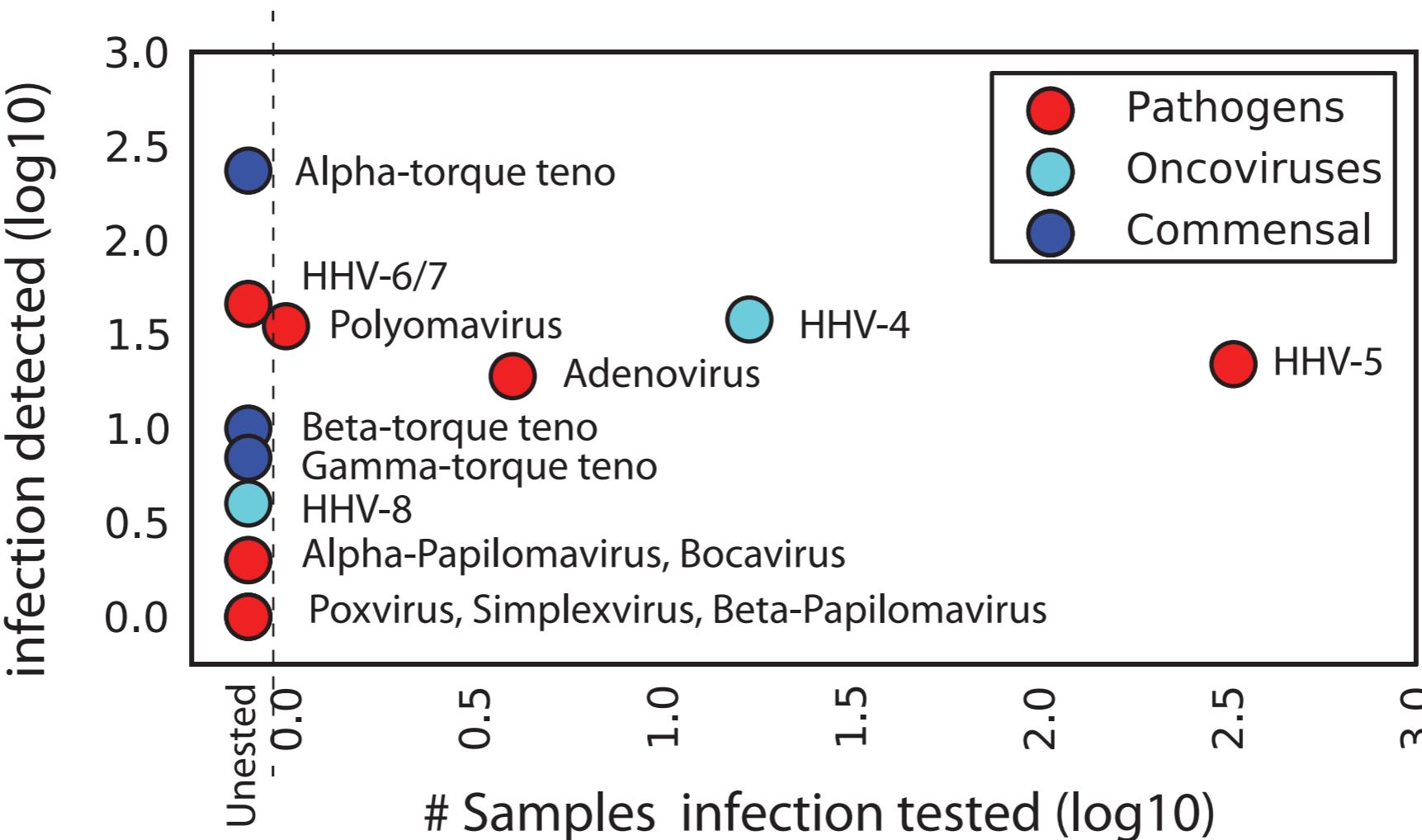
Show: 10

Search:

Name	Gen_Equ	Percentile
Candidatus Midichloria	0.28	99.516908
P2likeviruses	34.09	98.711755
Gammatorquevirus	169.43	98.711755
Escherichia	123.17	98.389694
Alkaliphilus	0.11	98.228663
Shigella	8.45	96.940419
Chelativorans	0.11	95.169082
Enterocytozoon	0.15	94.363929
Citrobacter	0.31	93.236715
Psychrobacter	0.39	92.914654

Un-tested / diagnosed infections.

Many potential pathogen that we detected are infrequently clinically tested for.



Undiagnosed cases of infection.

I6, Cause of death:
Respiratory failure.



Sorted infection data for I6.

Use back to return to cohort or dropdown menu above to switch between cohorts.

Show: 10 Search:

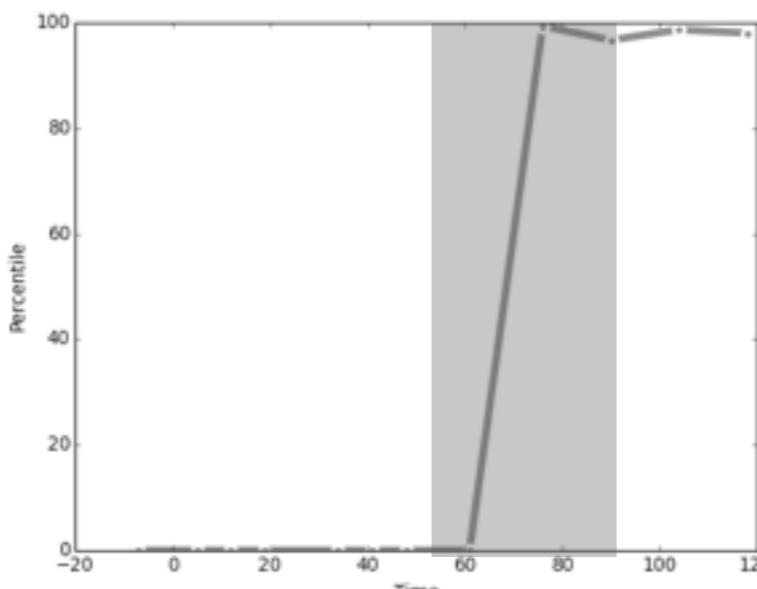
Name	I6_BC	I6_D-1	I6_W1	I6_W2	I6_W3	I6_W4	I6_W5	I6_W6	I6_W8	I6_W10	I6_W12	I6_W14	I6_W16
WU Polyomavirus	0.000000	0.000000	0.000000	0.000000	0.000000	0.000000	0.000000	0.000000	0.000000	99.337748	96.688742	98.875497	98.013245
Human herpesvirus 5	0.000000	75.496689	88.079470	78.145695	0.000000	90.066225	80.132450	89.403974	91.390728	0.000000	82.781457	0.000000	0.000000
Enterocytozoon bieneusi	0.000000	0.000000	90.728477	95.364238	94.039735	0.000000	96.026490	98.013245	98.675497	99.337748	94.701987	92.715232	0.000000



WU Polyomavirus
(Rare virus that causes severe respiratory infection)

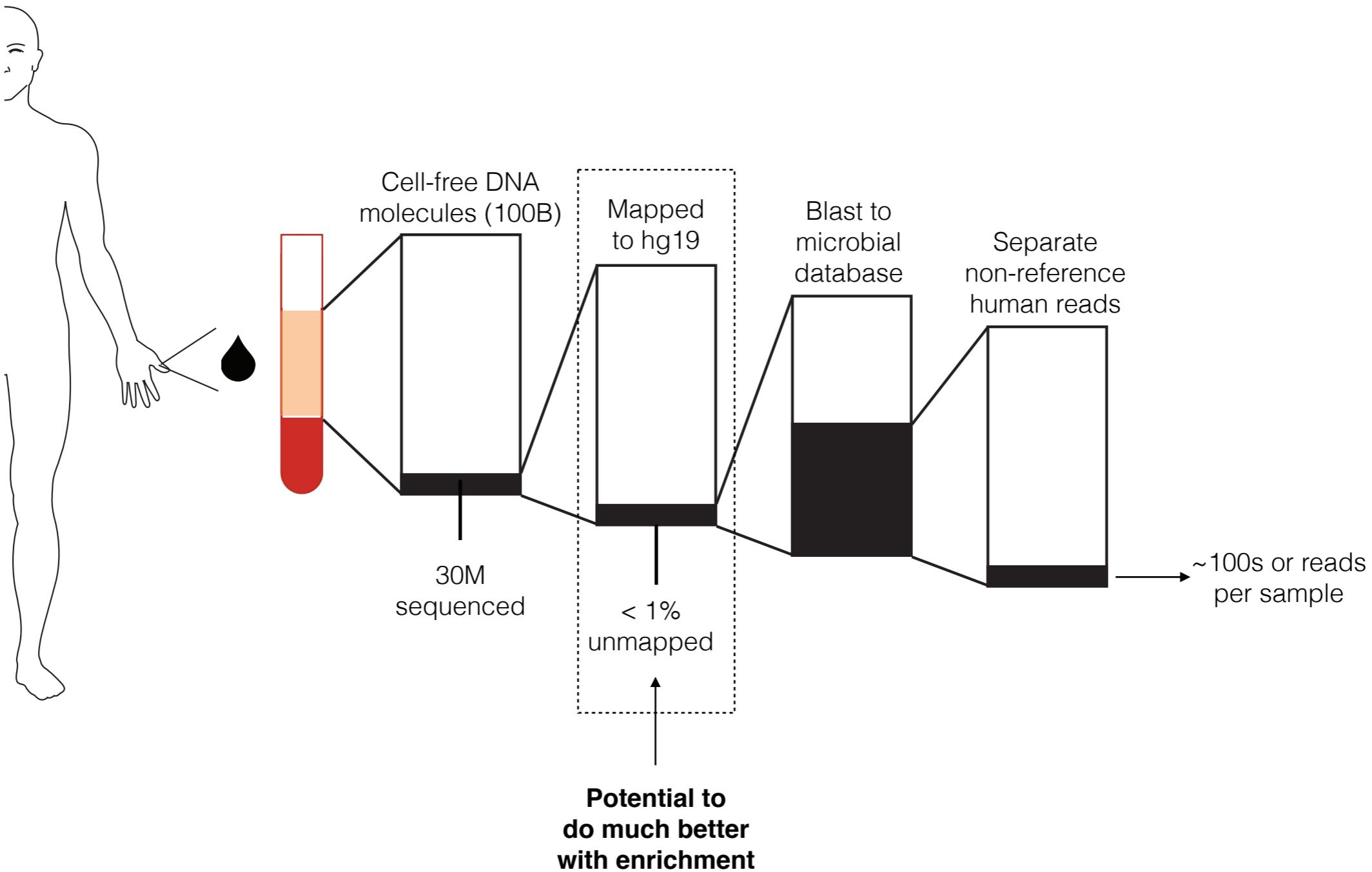


Very high load of WU Polyomavirus during time period of negative clinical test results.



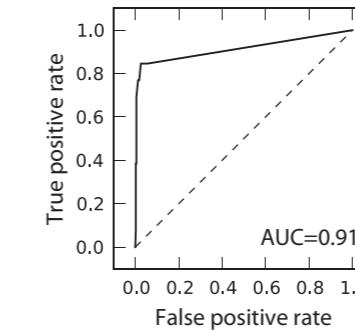
Multiple (-) tests for
BK polyomavirus

Also, headroom for improvement.

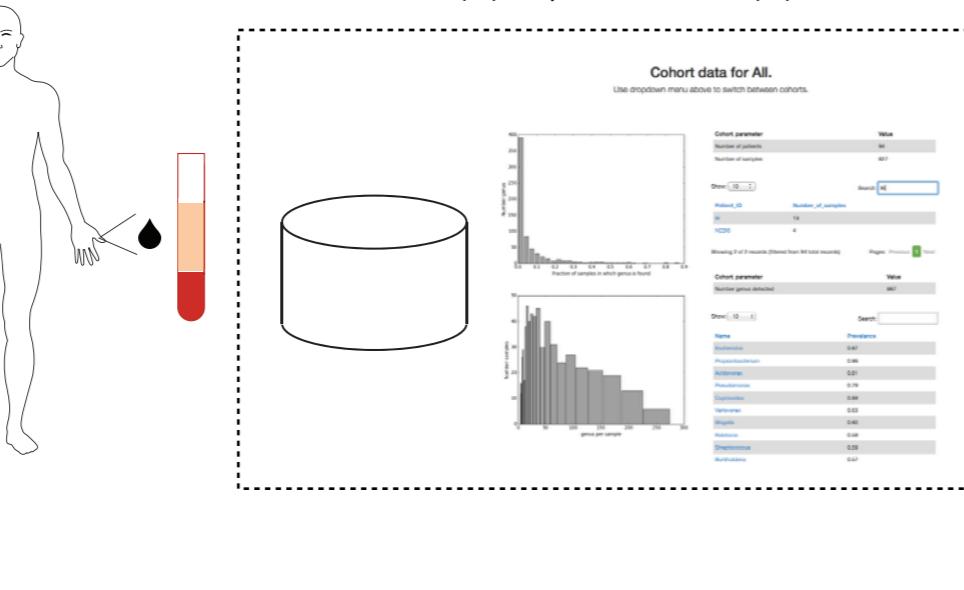


Infectious disease diagnostics from cell-free DNA.

(3) Clinical studies: Lung, BMT
In review, 2014



Infectome (1) Pipeline and (2) Browser



(4) Biopsy replacement
Stanford pathology

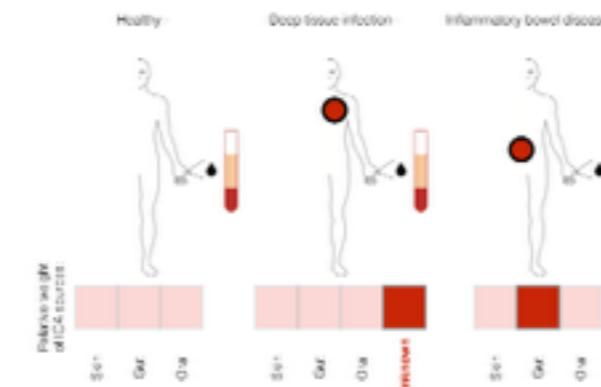


(5) Undiagnosed infections
Sandhya Kharbanda, Stanford BMT

A screenshot of a table titled 'Sorted infection data for I6'. It shows a list of infections with their prevalence across different samples. The table includes columns for Name, I6_B1, I6_D-1, I6_W1, I6_W2, I6_W3, I6_W4, I6_W5, I6_W6, I6_W7, I6_W8, I6_W9, I6_W10, I6_W11, I6_W12, I6_W13, I6_W14, and I6_W15. The top row shows WU Polyomavirus with a prevalence of 0.000000 across all samples. The bottom row shows Enterocysto-
zoon with a prevalence of 0.000000 across all samples.

Name	I6_B1	I6_D-1	I6_W1	I6_W2	I6_W3	I6_W4	I6_W5	I6_W6	I6_W7	I6_W8	I6_W9	I6_W10	I6_W11	I6_W12	I6_W13	I6_W14	I6_W15
WU Polyomavirus	0.000000	0.000000	0.000000	0.000000	0.000000	0.000000	0.000000	0.000000	0.000000	0.000000	0.000000	99.337748	96.686742	98.675497	99.013245		
Human herpesvirus	0.000000	75.496689	88.079470	78.145695	0.000000	90.066225	80.132450	89.403974	91.390728	0.000000	82.781457	0.000000					
Enterocysto- zoon	0.000000	0.000000	90.728477	95.364238	94.039735	0.000000	96.026490	98.675497	99.337748	94.701987	92.715232	0.000000					

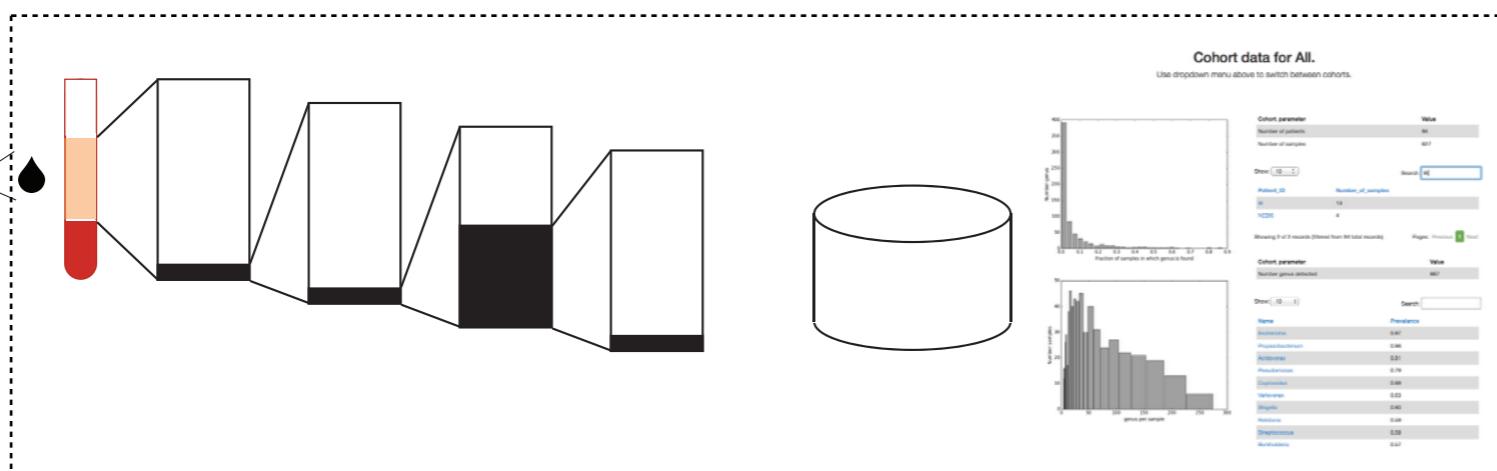
(6) Body site composition and anomalies
Relman lab, March of Dimes



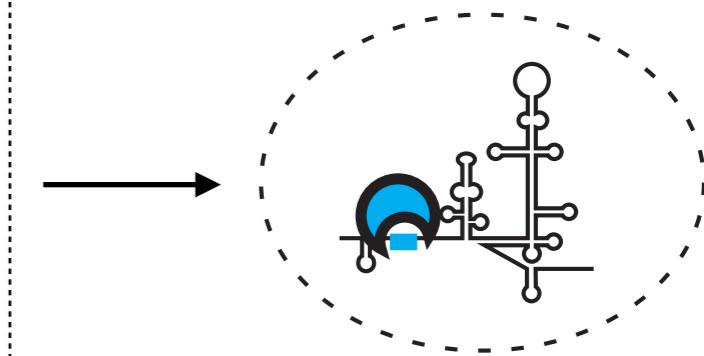
Sequencing also as a tool to study infection mechanism?



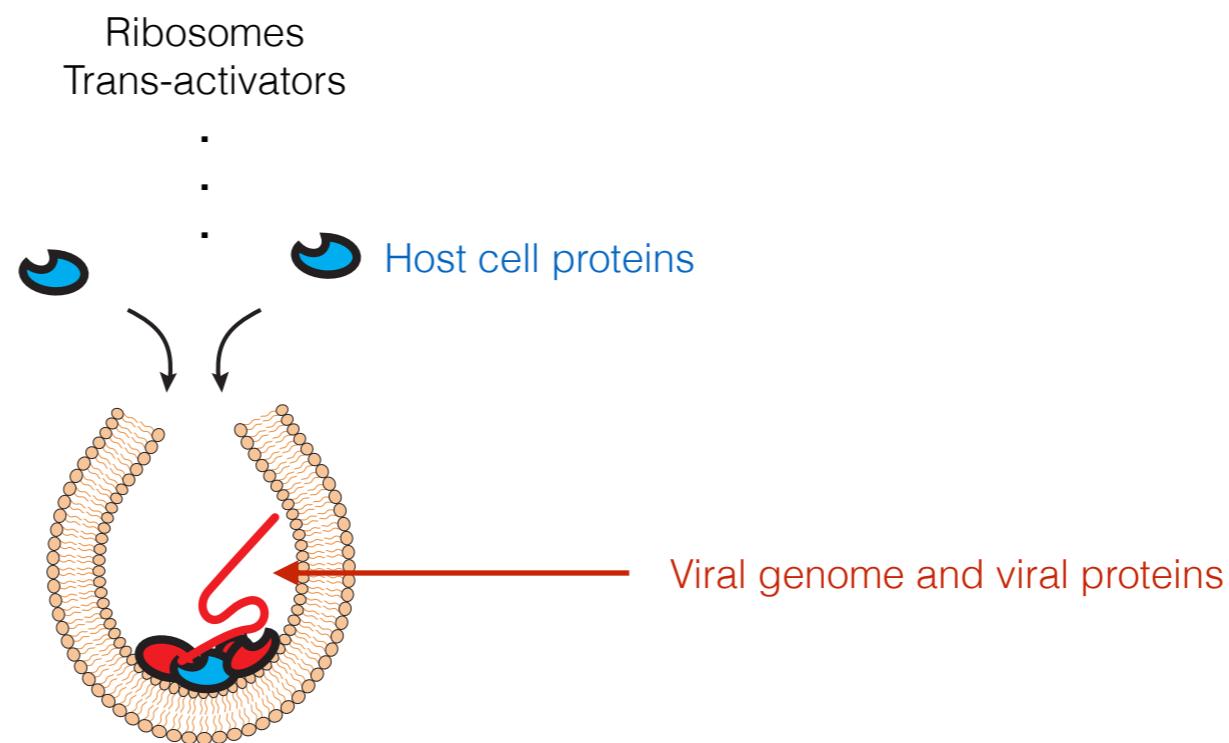
Sequencing as diagnostics -



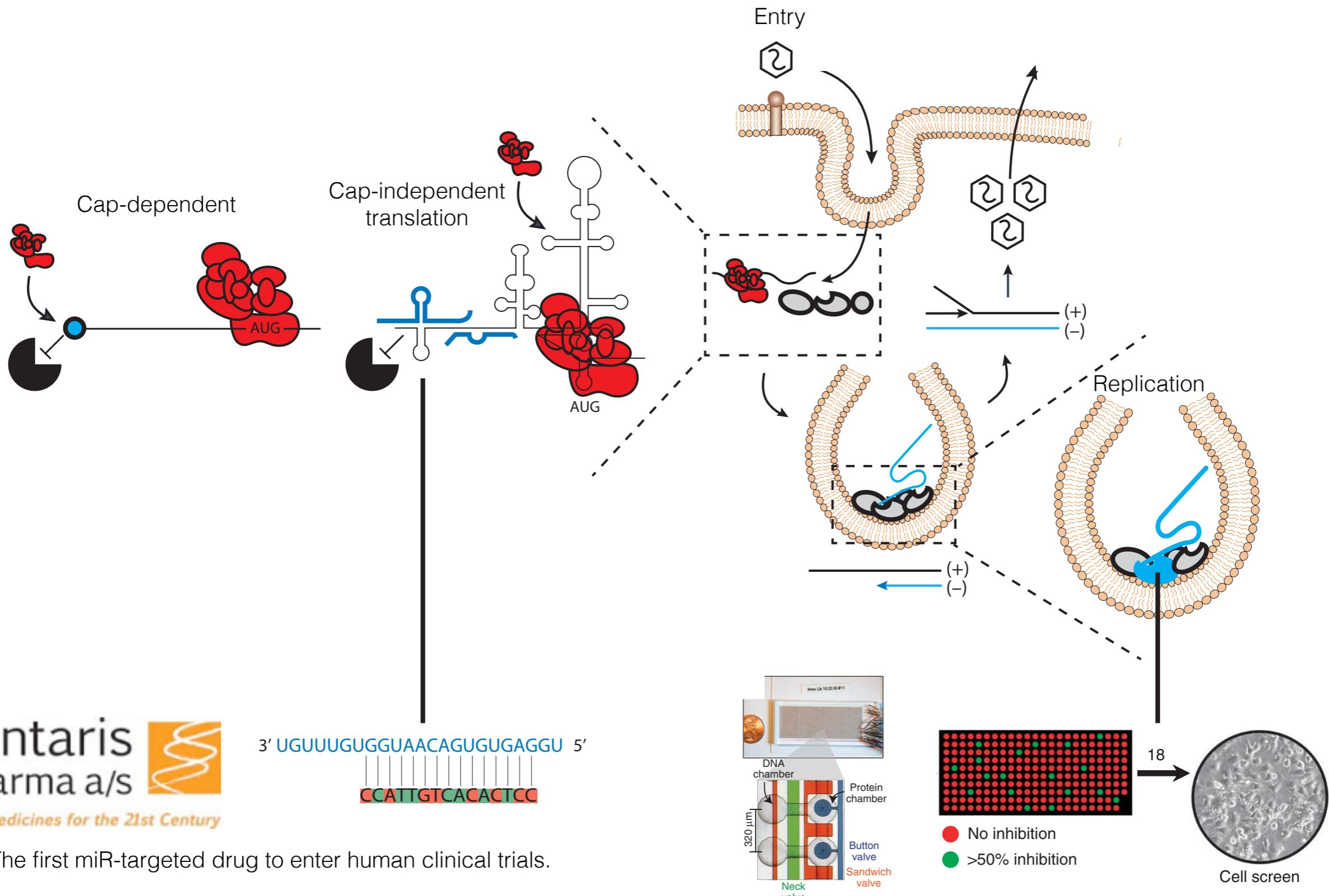
Mechanism relies on interactions -



Viral recruitment of cell host factors is critical.



Human host proteins required Hepatitis C virus lifecycle.

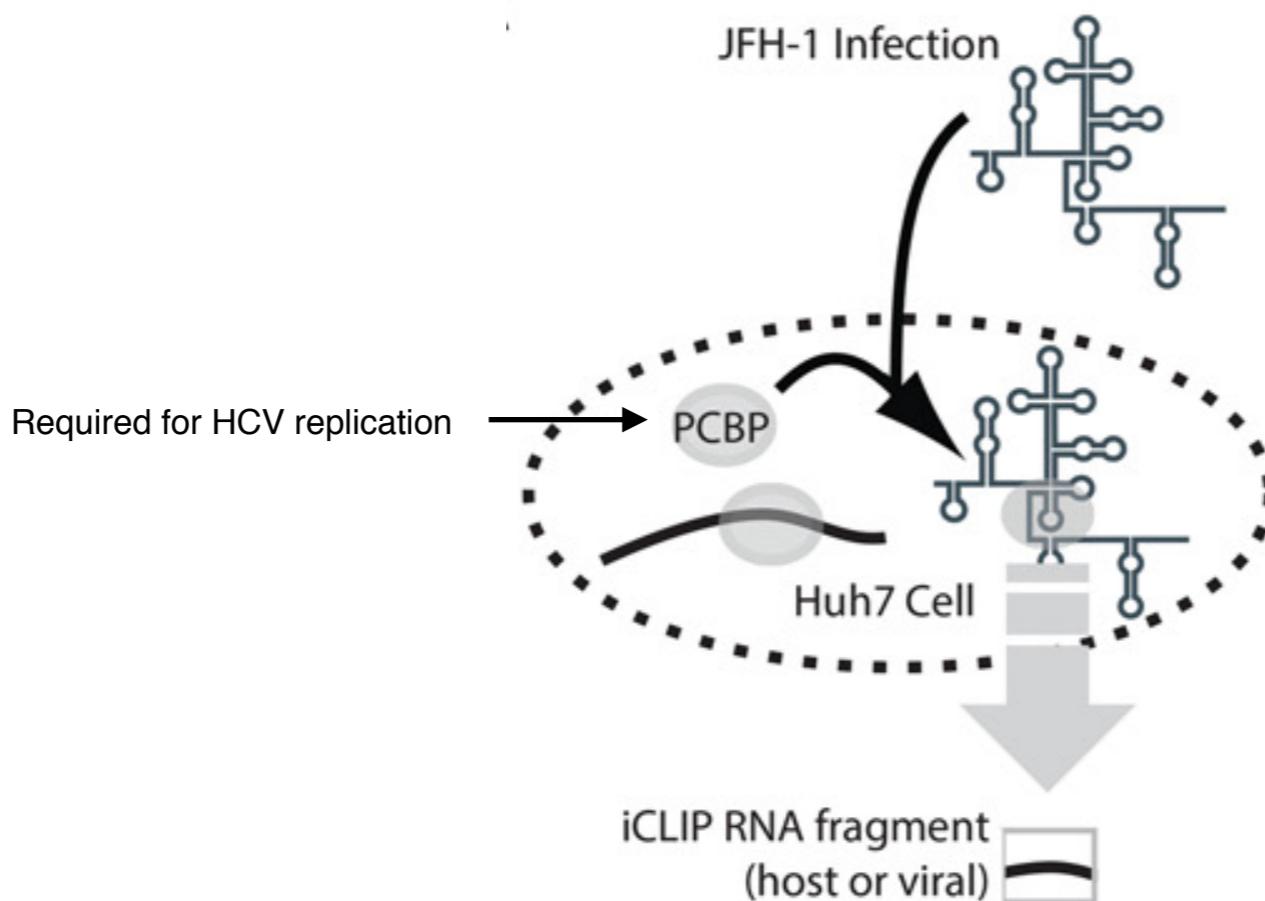


santaris
pharma a/s

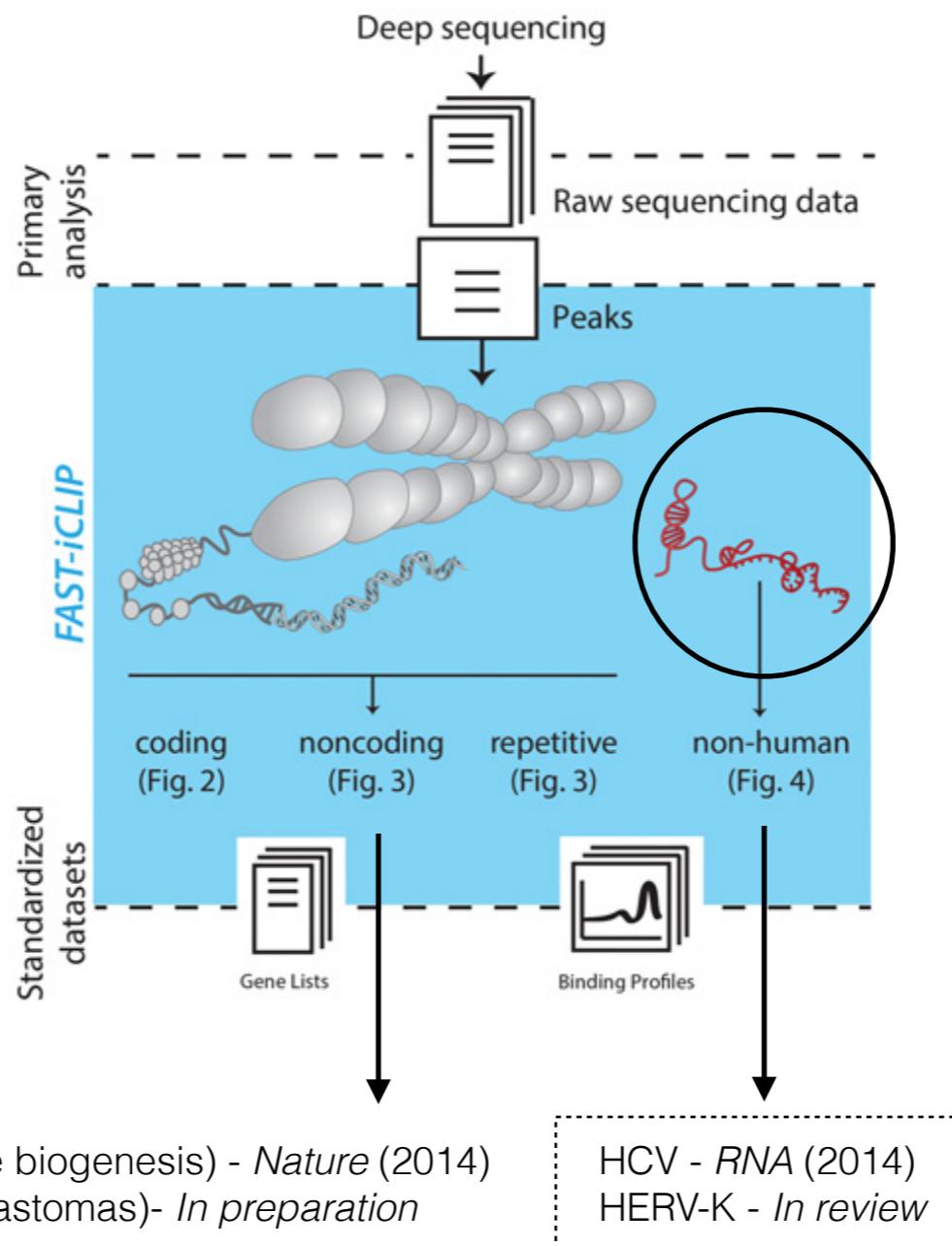
RNA Medicines for the 21st Century

The first miR-targeted drug to enter human clinical trials.

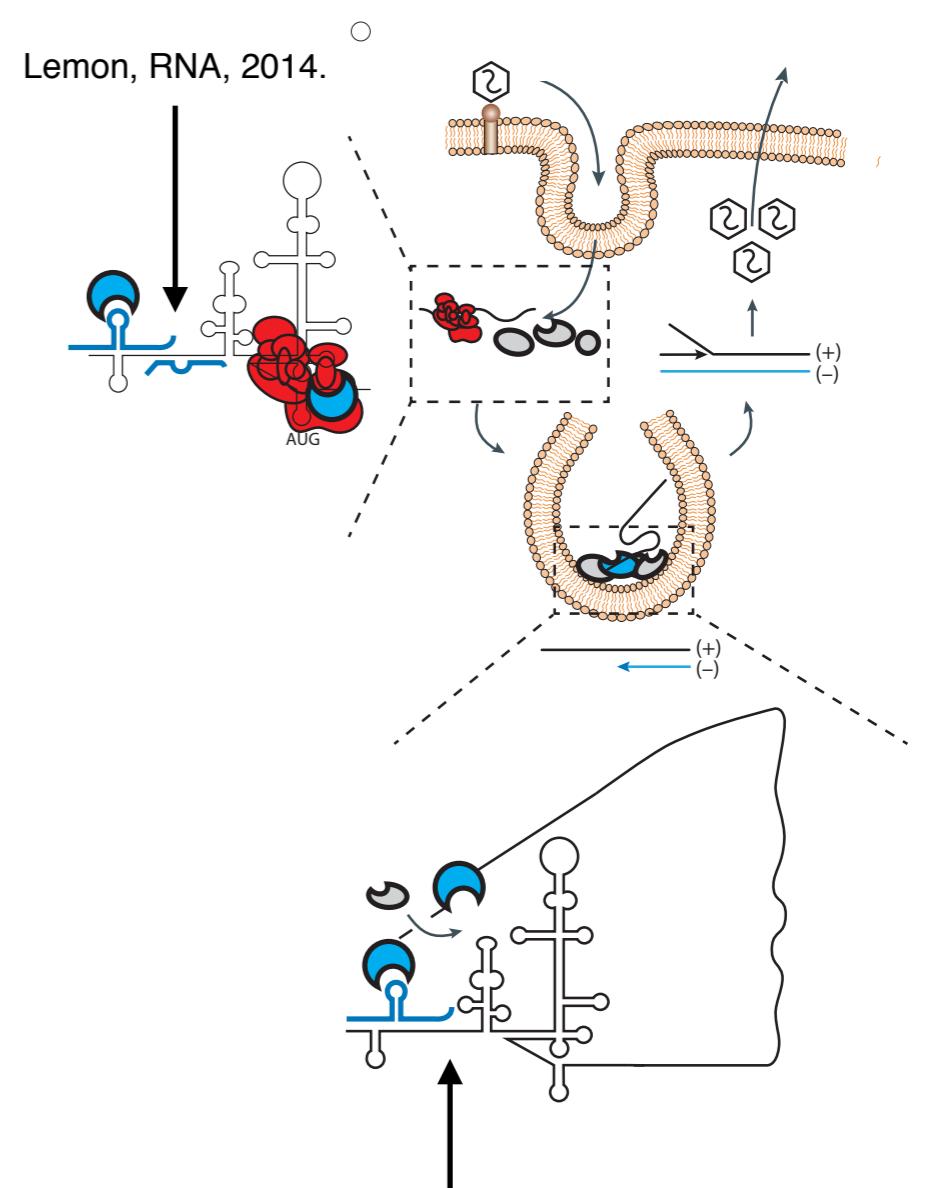
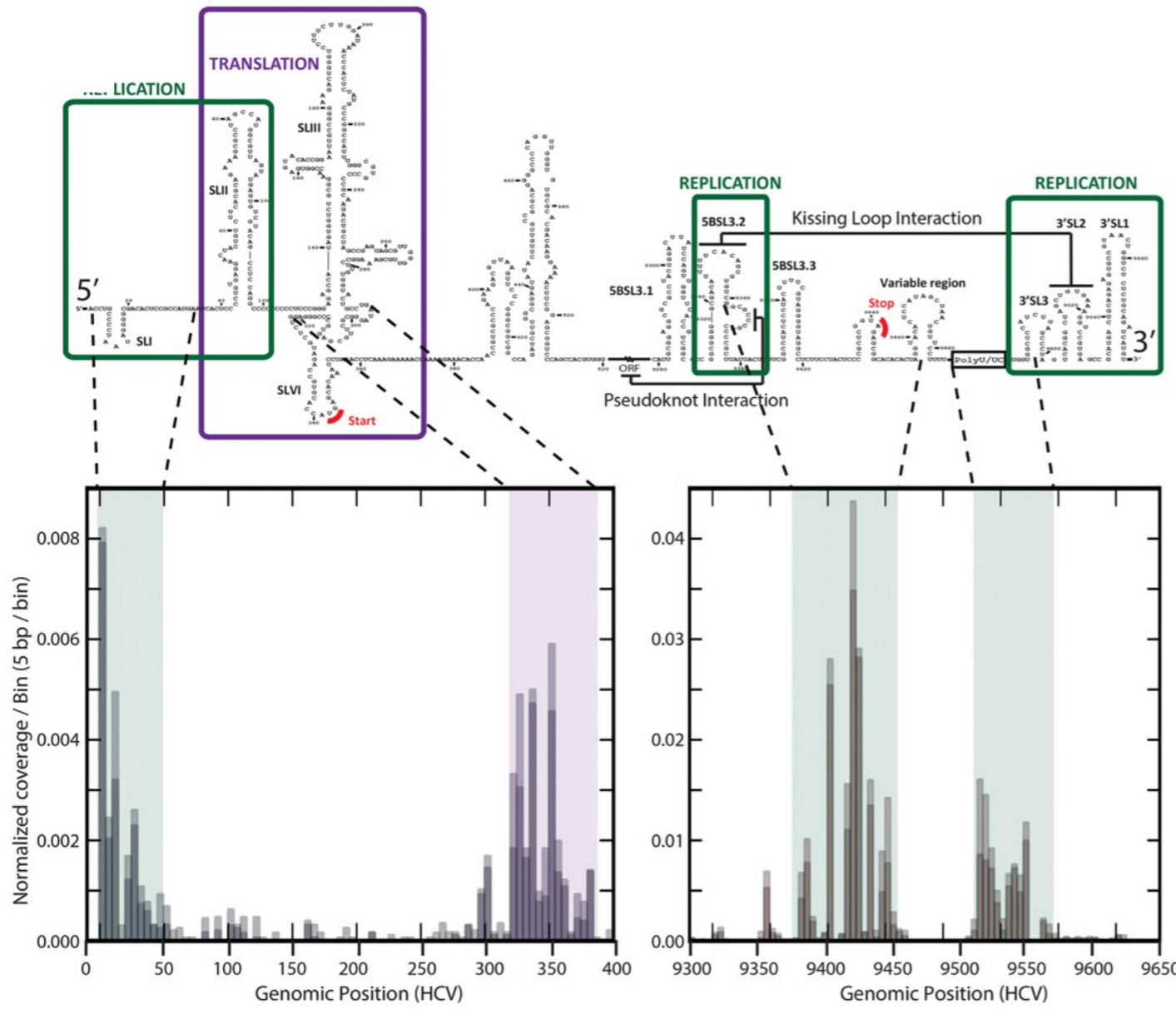
Apply to HCV viral-host factor interactome.



Pipeline for RNA-protein interactome of viruses.



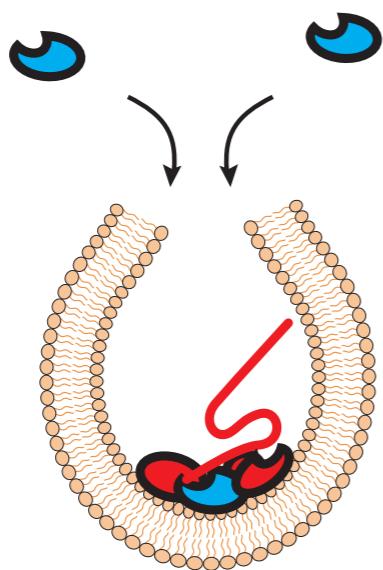
New insights about HCV lifecycle.



But, what about viral products interacting with host?

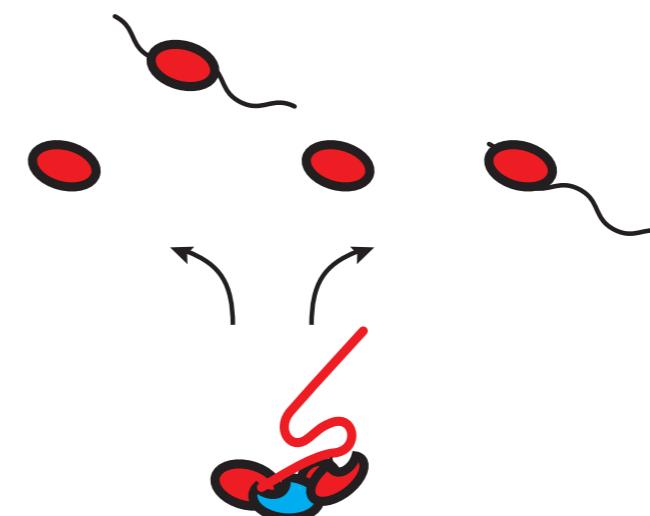
Host factor co-option -

- HIV
- Ebola
- HCV
- etc.

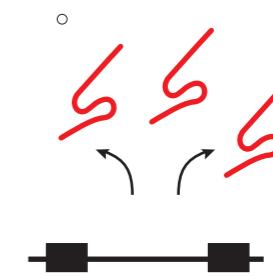
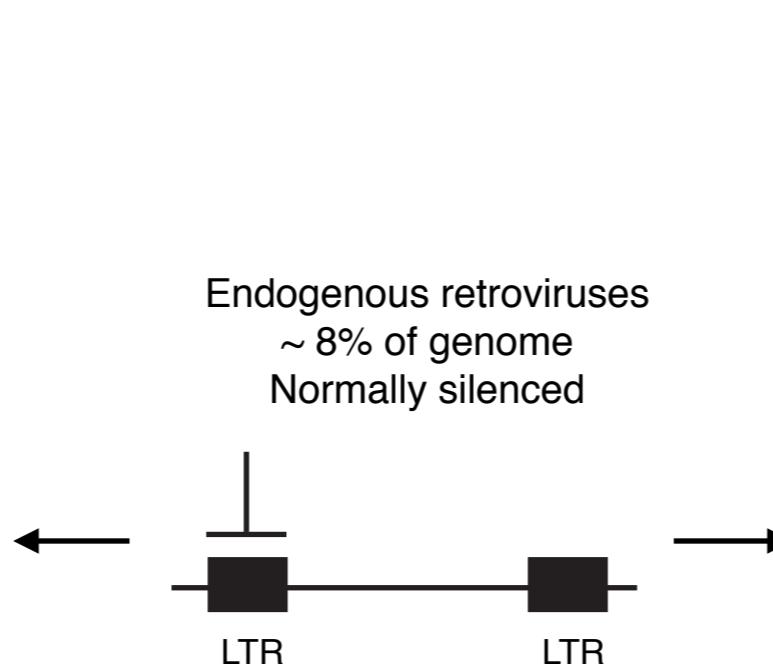
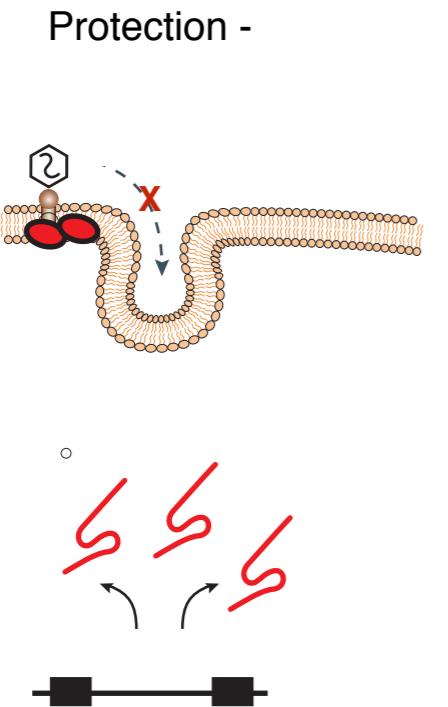


Viral protein interactome -

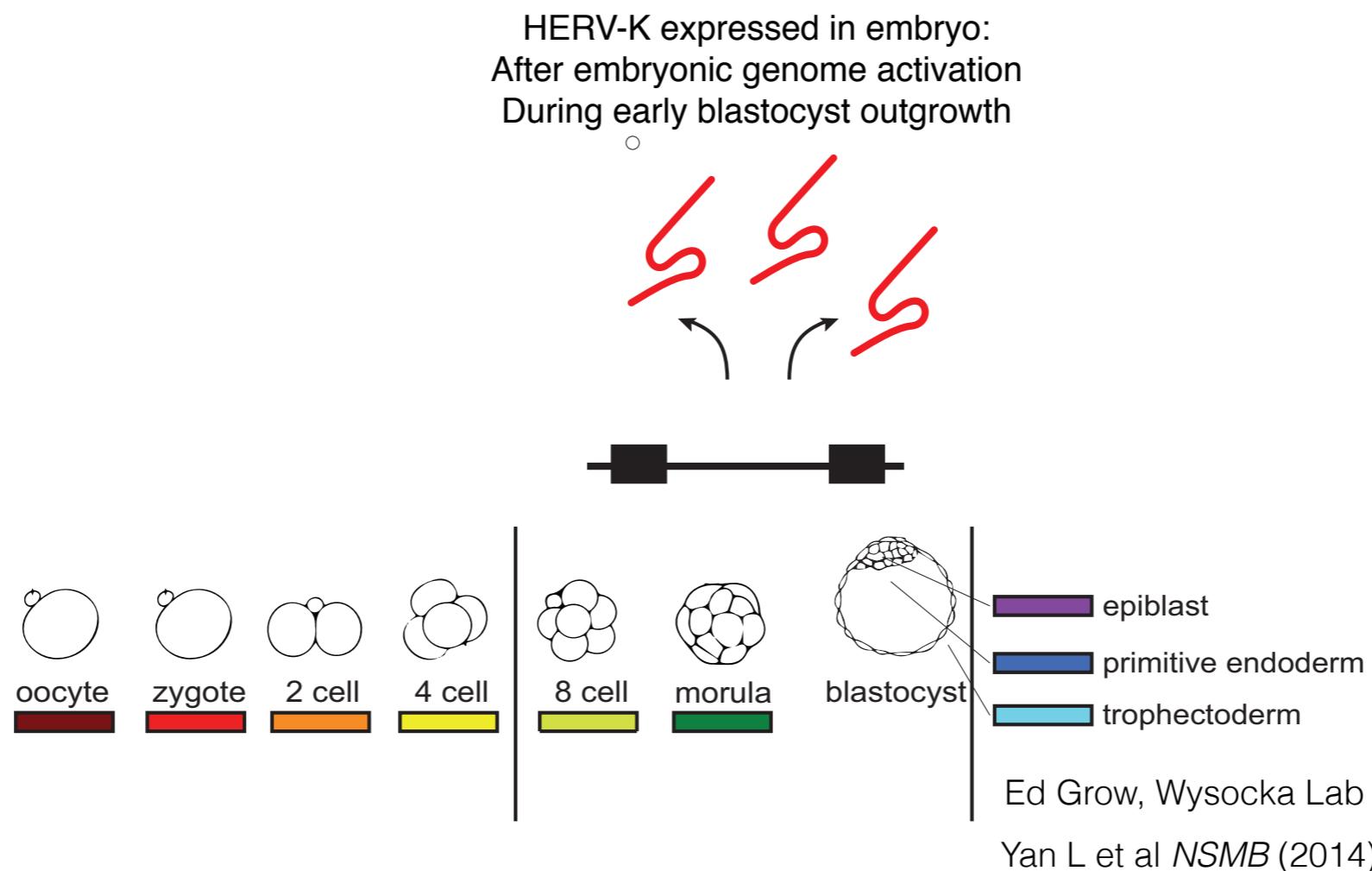
- Interaction with host?



Resurrection of endogenous retroviruses.

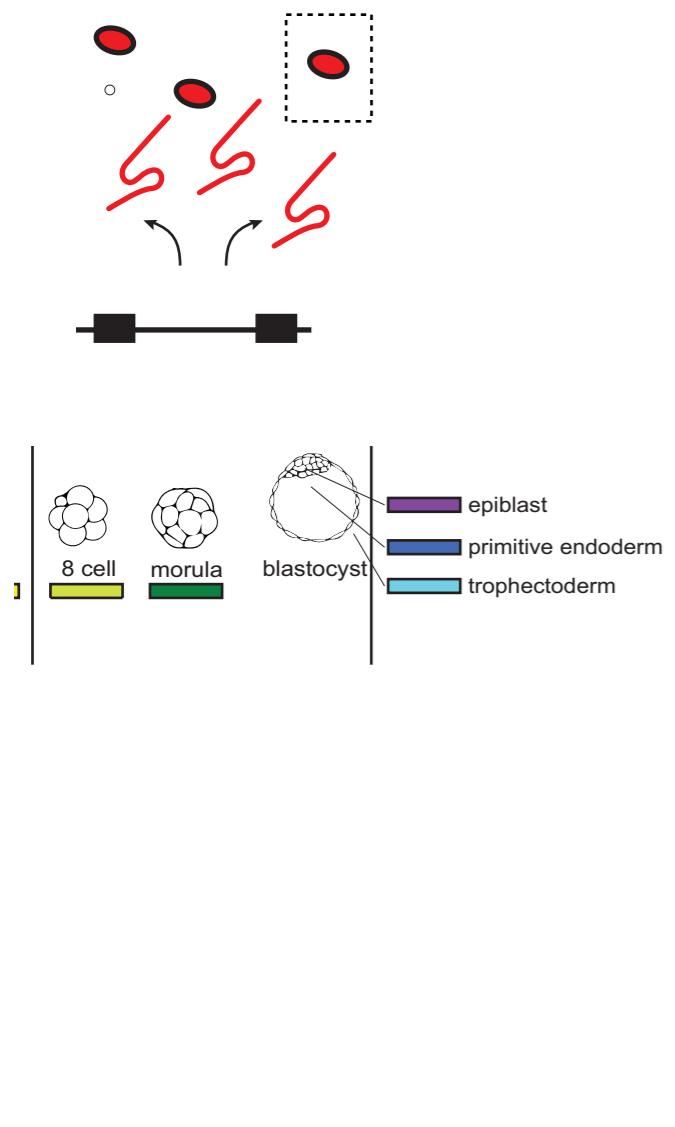


HERV-K retrovirus detected in signal cell embryo RNA-seq.

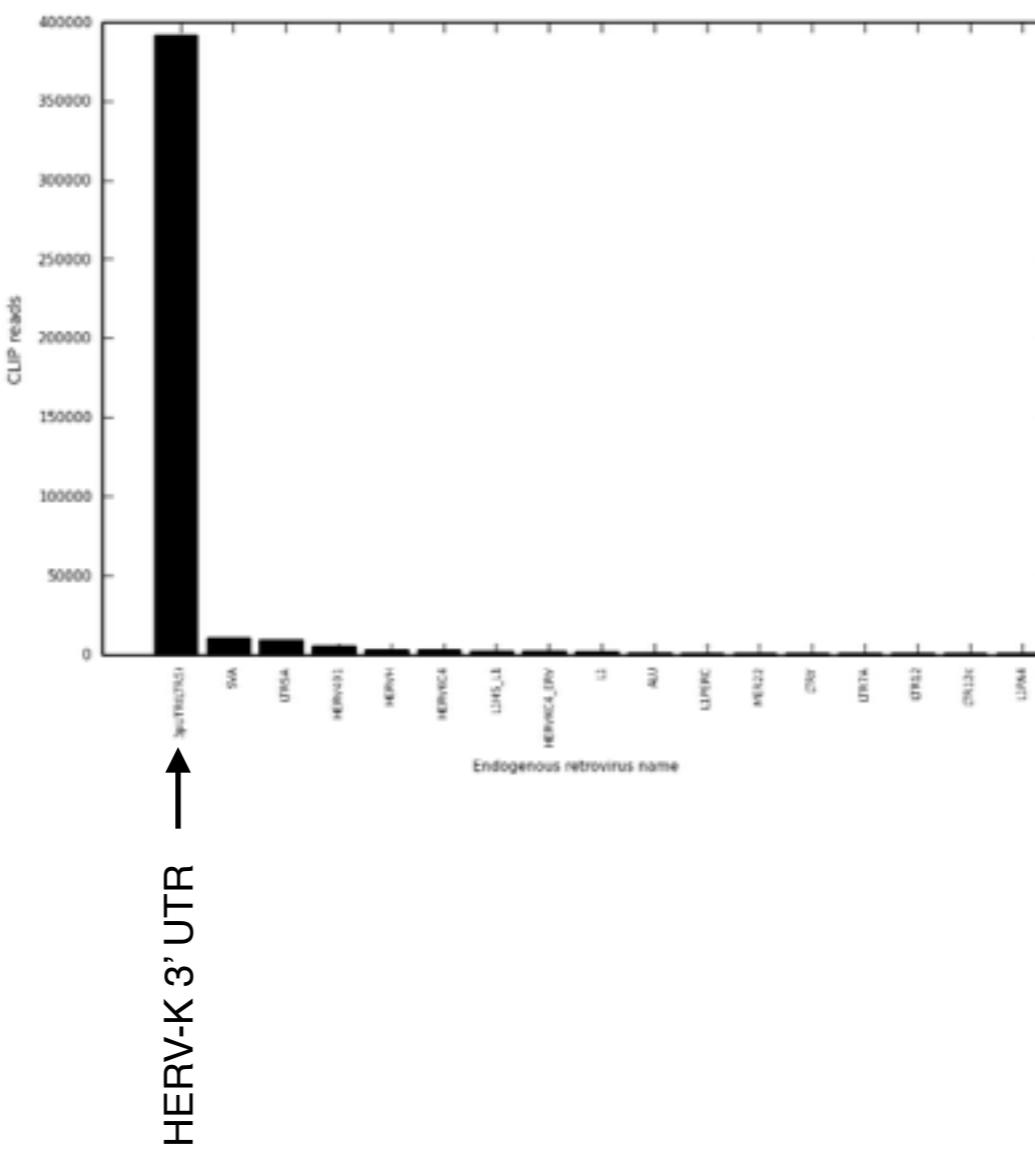


CLIP shows retroviral proteins active in embryo.

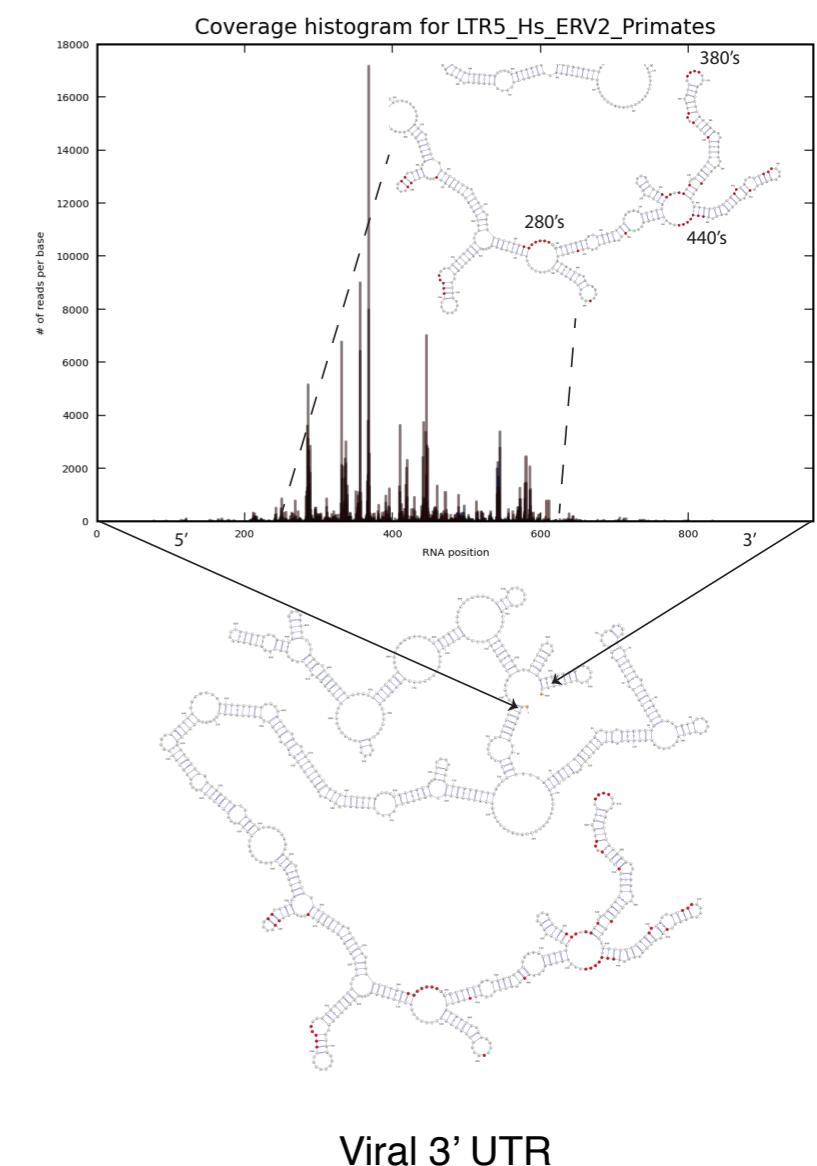
CLIP Rec, a viral protein
required for nuclear export in
embryonic carcinoma cells (hECCs)



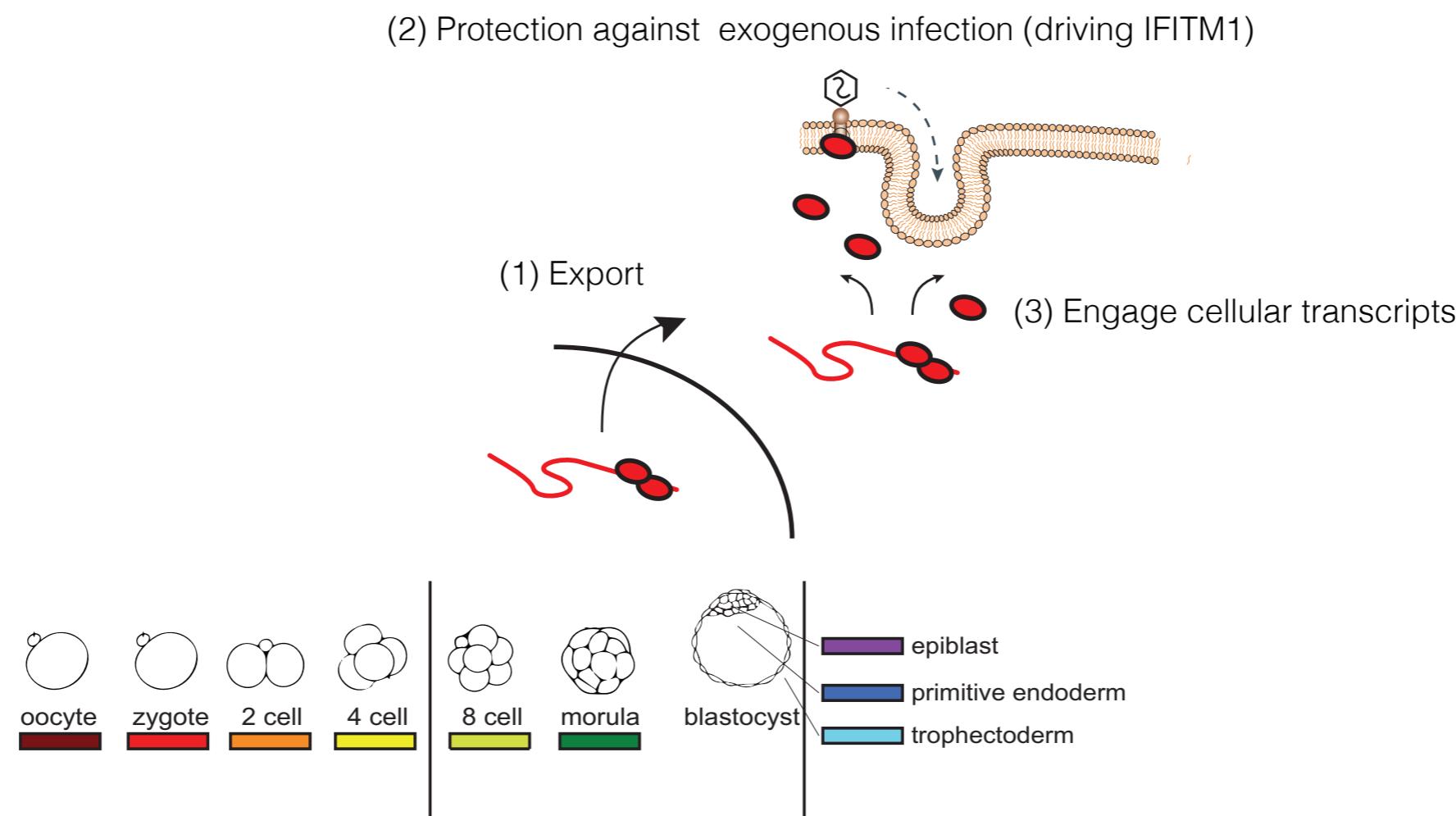
Read counts to regions in retroviral index -



Hits in Rec-responsive element region -
Lower et. al. PNAS (1993, 1996)



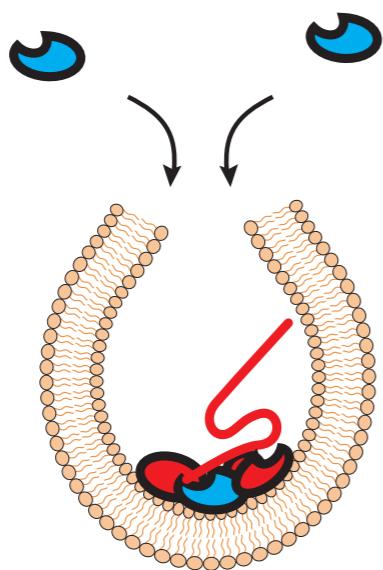
Retroviral proteins present in early human development.



CLIP is a useful tool for studying viral interactomes.

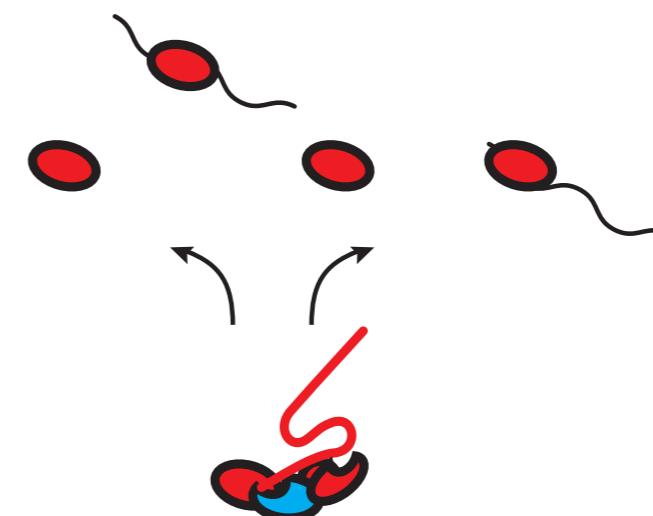
Host factor co-option -

- HIV
- Ebola
- HCV
- etc.



Viral protein interactome -

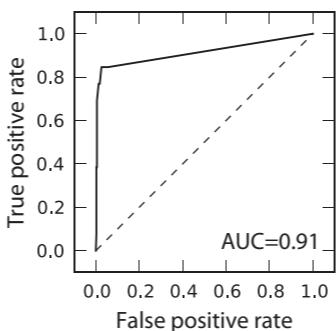
- Interaction with host
- Retroviruses



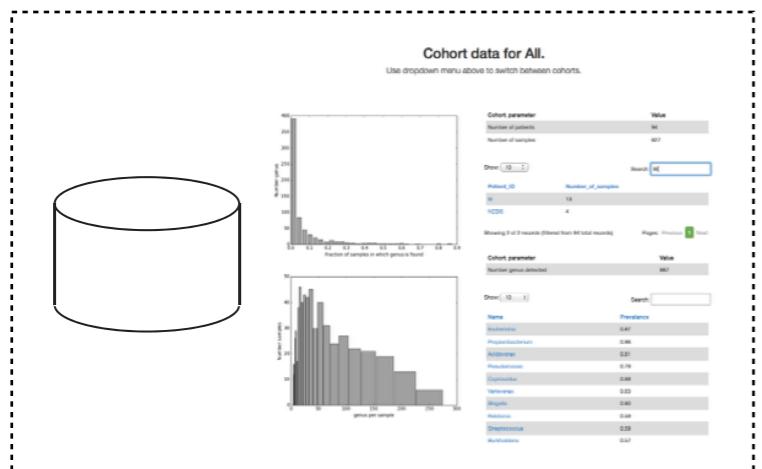
Diagnostics

Mechanism

(3) Clinical studies: Lung, BMT
In review, 2014



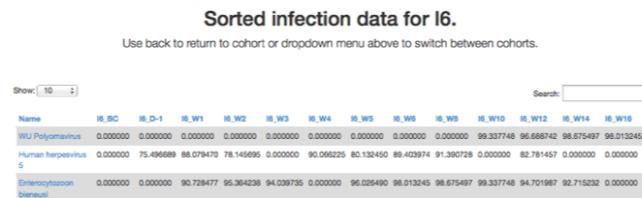
Infectome (1) Pipeline and (2) Browser



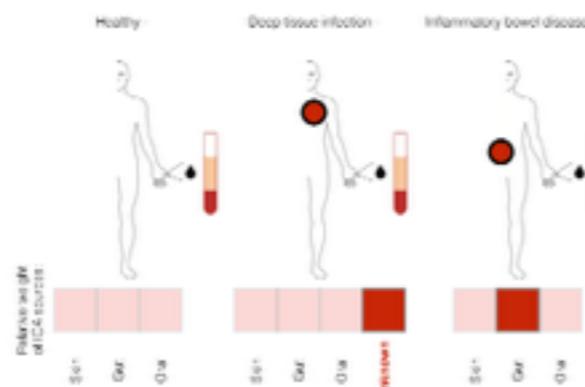
(4) Biopsy replacement
Stanford pathology



(5) Undiagnosed infections
Sandhya Kharbanda, Stanford BMT

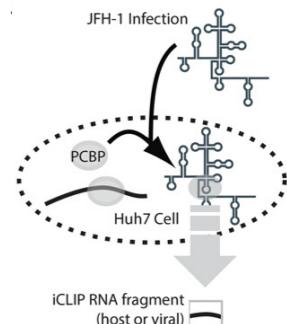


(6) Body site composition and anomalies
Relman lab, March of Dimes



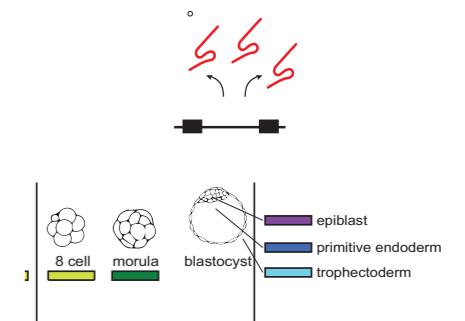
(8) Human
-Nature, 2014
- In preparation
(Cho, Doudna Labs)

(9) Viral genomes
RNA, 2014

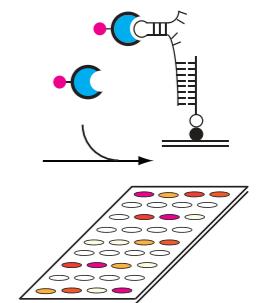


(7) CLIP pipeline
RNA, 2014

(10) Retroviral genomes
In review, 2014



(11) Validation tools
Nature Methods, 2012

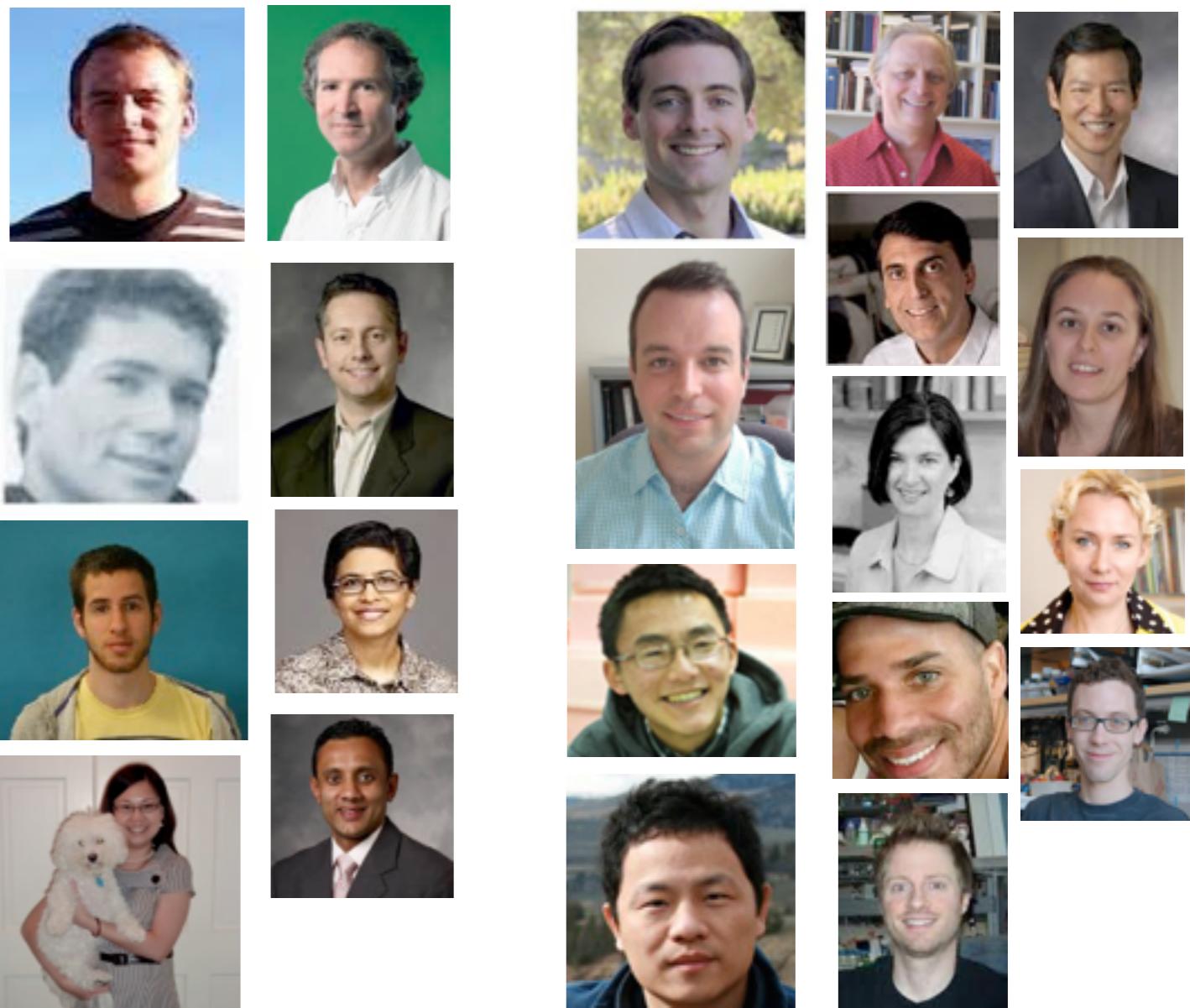


Thanks

Funding



People



+ Quake and Chang labs, Sequencing Core