Cure – the problem we solve for, challenges we face and progress to date

Stephen Becker, MD
Chief Scientific and Medical Officer
BryoLogyx Inc.
What problem are we solving: what motivates “cure” research?

• Lifelong ART is a challenge and may be associated with adverse events, long-term safety considerations, drug resistance, adherence challenges and QOL issues [patient centered]
What problem are we solving: what motivates “cure” research?

• Expanding ART to those in need and the infrastructure needed for treatment and care is not remotely sustainable in the current, flat-funded, environment [public health and economic centered]

**Number of PLWA on ART**

<table>
<thead>
<tr>
<th>Year</th>
<th>People living with HIV</th>
<th>People receiving treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>2000</td>
<td>28.9 million</td>
<td>770,000</td>
</tr>
<tr>
<td>2005</td>
<td>31.8 million</td>
<td>2.2 million</td>
</tr>
<tr>
<td>2010</td>
<td>33.3 million</td>
<td>7.5 million</td>
</tr>
<tr>
<td>2012</td>
<td>34.5 million</td>
<td>11 million</td>
</tr>
<tr>
<td>2013</td>
<td>35.2 million</td>
<td>13 million</td>
</tr>
<tr>
<td>2014</td>
<td>35.9 million</td>
<td>15 million</td>
</tr>
<tr>
<td>2015</td>
<td>36.7 million</td>
<td>17 million</td>
</tr>
<tr>
<td>2016</td>
<td>36.7 million</td>
<td>19.5 million</td>
</tr>
</tbody>
</table>

**Global resource allocation and gap**

Source: UNAIDS Data 2017
What problem are we solving: what motivates “cure” research?

• Incidence rates globally have flattened; in the setting of the projected ‘population bulge’...largest at risk population since the beginning of the epidemic [public health centered]
Truly transformative approaches are needed

• For those already HIV-infected → drug, product form and delivery of care innovation: might a functional cure (sustained remission off ART) be possible?
• For those not HIV-infected → preventative vaccination

NEW PARADIGM AHEAD?
What cure must accomplish; what kills cure

**What cure must accomplish**

- Overcome daunting biology
- Validation of a biomarker
- Achievable TPP
- Demonstration of cost-effectiveness
- Engagement by industry
What cure must accomplish; what kills cure

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**What ends cure**
- Absence of a biomarker kills product development
- Transmission during treatment interruption
- Emergence of ART drug resistance
- Induction of oncogenes
- Unacceptable adverse events
Disclosures

• Serving as Chief Scientific and Medical Officer for BryoLogyx Inc.
• The “ART counterfactual” is compelling
• I am agnostic to the question of whether cure is possible; the data will inform us
• The cure field is in need of a positive signal in order to justify funding at the current level (HIV/AIDS exceptionalism no longer applies)
Topics

• The components of cure
• Case studies, work to date and the natural history that informs cure research
• Defining the HIV reservoirs
  – Anatomic and cellular heterogeneity
  – Clonal expansion
  – HIV integration sites
• Measuring the HIV reservoir (biomarker central)
• Secondary lymphoid organs – the B-cell follicle
• Final thoughts
The components of cure
The elements of an HIV cure - the agreed approach led by an NIH vision

- Viral suppression and immune protection
- HIV reservoir depleted
- Immune system mediated viral killing

Kick → Kill

Modified from Chun, et. al., Nature Immunol 2016
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- **Viral suppression and immune protection**
  - Early ART initiation

- **HIV reservoir depleted**
  - Latency reversal; reduced reservoir size

- **Immune system mediated viral killing**

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- Discontinuation of ART is permitted in the setting of depleted latent HIV reservoirs and restored immune function; sustained virologic remission, and immune stability, persist in absence of therapy

Modified from Chun, et. al., Nature Immunol 2016
Case studies, early experience and natural history informing cure
Time to viral rebound following ART

1 log variability similar to other studies
Time to viral rebound following ATI

- Typical rebound after ART cessation

Viral load

ART

Time in remission

2 3 8 10 28

Months

2

Years

7 12 Ongoing

Time to viral rebound following ATI

- Typical rebound after ART cessation
- Boston patients: Method - Stem cell transplants
- Mississippi baby: Method - Early ART initiation
- Visconti cohort/post treatment controllers: Method - Early ART initiation
- French teenager: Method - Early ART initiation
- Mayo clinic patient: Method - Stem cell transplant
- The Berlin Patient (Timothy Brown): Method - Chemotherapy + 2 full stem cell transplants

All started ART early after infection.
Earliest ART does not prevent reservoir formation

Individual in PrEP program who began cART 10 days following infection; HIV suppression for 34 mos

Early ART resulted in reduced reservoir size and seeding

# Elite and post-treatment controllers – experiments of nature in HIV

<table>
<thead>
<tr>
<th>Attribute</th>
<th>Elite controller</th>
<th>Post-treatment controller</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frequency</td>
<td>2.5-5.0%</td>
<td>5-15%</td>
</tr>
<tr>
<td>HLA type</td>
<td>Protective</td>
<td>Not protective</td>
</tr>
<tr>
<td>HIV RNA level at infection</td>
<td>Low</td>
<td>High</td>
</tr>
<tr>
<td>HIV-specific CD8 responses</td>
<td>Strong</td>
<td>Weak</td>
</tr>
<tr>
<td>CD8 (immune) activation</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>Symptomatic at diagnosis</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>ART initiation</td>
<td>No ART needed</td>
<td>Begun in acute HIV infection stage</td>
</tr>
</tbody>
</table>
What did we learn?

• In the majority of cases viral load rebound occurs with days to 4-6 weeks following ART interruption; this is a considerable variability

• There exists a unique group of post-treatment controllers, distinct from elite controllers, and they may hold understandings to cure

• ART within 10-12 days of infection will not prevent formation of the latent reservoir

• ART type does not influence the time to decay of latently infected cells
The HIV reservoirs – insights
The reservoir – heterogeneous cellular subsets and anatomic sites
Reservoir decay does not vary by era of ART

$t_{1/2} = 3.7$ years

Finzi, 1999; Siliciano, 2003 NatMed
Crook, JID 2015
Viral composition of the reservoir

Resting, latently infected CD4 cells

Barton, Trends Micro 2016
Viral composition of the reservoir

Mitogen stimulation: induced and non-induced proviruses
Viral composition of the reservoir

Non-induced virus can be intact and competent or defective (hypermutated, internal deletion, stop codon, etc).

Barton, Trends Micro 2016
Viral composition of the reservoir

Additional rounds of stimulation / activation will induce cells containing replication-competent virus

Barton, Trends Micro 2016
Reservoir measurement
Measuring what matters

- Assays and the cells measured -

Total HIV DNA

Integrated HIV DNA

Cell-associated HIV RNA

Cell-associated HIV provirus

Cells releasing HIV virions

Replication-competent HIV
What comprises the latent reservoir?

- Siliciano spherical representation of the reservoir
- PCR-based assays
- Intact virus assays
- Viral outgrowth assay

Modified Bruner, CROI 2018
What comprises the latent reservoir?

- Intact
- Hypermutated
- Hypermutated & Deleted
- Deleted

Bruner, in press
Improved measurement of the latent reservoir

Bruner, in press
Improved measurement of the latent reservoir

Bruner, in press
What did we learn?

• Intact and replication competent virus is what we want to measure; defective and mutated variants are not relevant as a biomarker
• Repeated rounds of stimulation will be required to quantify the viral population of interest
• PCR-based measurements of the reservoir should be abandoned [tell this to our Europeans colleagues]
• Digital drop intact proviral DNA (IPDA) is the preferred and optimized assay; clinical correlation is ongoing
The establishment of latency and the maintenance of HIV persistence – evidence of a highly adapted virus
Phases of viremic decay following ART

I. Death of activated CD4 cells; \( t_{1/2} \) of 1-2 days

II. Death of partially activated CD4 cells; tissue macrophages and dendritic cells; \( t_{1/2} \) of 14 days

III. Slow depletion of resting CD4 cells; \( t_{1/2} \) of 44 months

IV. Clonal expansion

Adapted, Dahl, Antiviral Res, 2010
HIV integration – it’s not all the same

Composition of the pre-integration complex (PIC) can vary and determine nuclear import and thus proximity to host genome structures.
HIV integration – it’s not all the same

Composition of the pre-integration complex (PIC) can vary and determine nuclear import and thus proximity to host genome structures.

Site of integration is not random, but rather is biased in favor of genes that regulate cell growth.

Maldarelli, JCI, 2016
Clonal expansion during suppressive ART

Hughes, Cell Host and Microbe, 2016; Maldarelli, JCI, 2016
Clonal expansion is not random, but rather favors clones with biologic advantage.
Clonal expansion during suppressive ART

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Expanded clones carry replication-competent virus; ART does not prevent clonal expansion.

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Clonal expansion during suppressive ART

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Persistent clones may be activated and are responsible for residual viremia

Hughes, Cell Host and Microbe, 2016; Maldarelli, JCI, 2016
Clonal expansion during suppressive ART

Mechanisms of clonal expansion

→ homeostatic proliferation
→ interaction with antigen
→ integration site-dependent proliferation

Hughes, Cell Host and Microbe, 2016; Maldarelli, JCI, 2016
Reservoir formation and maintenance

Persistence → Latency → Inflammation → Homeostatic expansion → Persistence
Reservoir formation and maintenance

- **Suppression of viral replication** through tat and P-TEFb; Epigenetic silencing
- **Persistence**
  - Further suppression of viral transcription factors and cell silencing
- **Latency**
- **Inflammation**
  - Further suppression of viral transcription factors and cell silencing
- **Homeostatic expansion**
  - Clonal expansion; integration site biases
- **Cellular proliferation and exhaustion**
What did we learn?

• HIV integration into the host genome is not random, and favors sites of cell growth regulation
• Low level viremia on ART may be due to constitutive factors, immune activation, privileged anatomic sites and possibly antiretroviral pharmacology
• Expanded clones contain replication competent virus; suppressive ART does not prevent clonal expansion
• Clonal expansion maintains viral persistence
• This is a problem
The lymph node follicle – realizing differing human and simian pathobiology (and the cardinal efforts of Liz Connick)
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Secondary lymphoid organs in cure quantitative pharmacology’s delight and cure’s dilemma

Lymph node architecture
The secondary lymphoid organs are the major HIV reservoir in humans. The secondary lymphoid organs contain the largest proportion of HIV RNA+ cells, and is the predominant source of viral replication during ART. 

Modified from Bronnimann, 2018
The secondary lymphoid organs are the major HIV reservoir in humans

The B-cell follicle is a privileged site from CTL control; CD8 cells lack the CXCR5 homing receptor for entry.

Modified from Bronnimann, 2018
The secondary lymphoid organs are the major HIV reservoir in humans.

Dendritic cell trafficking provides a high and continuous concentration of virus, even under ART.
The secondary lymphoid organs are the major HIV reservoir in humans

Follicular CD4 cells are the most permissive to HIV infection

Modified from Bronnimann, 2018
What did we learn?

• Anatomic, viral and immunological factors enable new cellular infection in a microenvironment that favors viral replication and persistence
• Highly permissive to infection, follicular CD4 cells are relatively shielded from CTL responses
• If ART lymphatic pharmacologic insufficiency is real, we’d best be aware
• Immune-tropic therapeutic agents must specifically target the secondary lymphoid compartment
• This is a problem
So where does all this leave us?
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  - Immune modulation

Early ART may limit reservoir size and prolong time to viral rebound, but it does not prevent the establishment of persistence

Modified from Chun, et. al., Nature Immunol 2016
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HDACis induce viral transcription but do not reduce reservoir size; PKC modulators in preclinical eval, but with concerns re TI

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Improving HIV-specific responses is felt essential; no product has yet achieved this at a clinically meaningful level

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The brightest light; several agents with broad immunotherapeutic effect; breadth, potency, baseline sensitive, PK and cost issues pertain

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Perhaps our best hope to address the privileged lymphoid compartment

Modified from Chun, et. al., Nature Immunol 2016
Final thoughts

• The biology of latency, persistence and the heterogeneity of the reservoir is more complex than initially thought
• Secondary lymphoid organs are the major site of the reservoir in humans; overcoming viral and immune dynamics may be key to cure
• A validated biomarker of reservoir size, (predictive of time to viral rebound) is critical, and will likely emerge as the singular factor that permits, or dissuades industry engagement; cure will not happen without industry participation
Final thoughts

• The kick and kill paradigm is sound, but is a blunt instrument where surgical precision is needed

• We must understand the problem we solve for - the need for transformative approaches is critical → We are not winning this war

• To succeed cure must demonstrate that sustained remission off ART is possible, cost-effective, advantageous, and desired by end users

• Considering cure as a public health imperative, if we are to fail, we must fail fast, and integrate lessons for tomorrow