DISCLOSURE

I have no relevant commercial relationships to disclose.
Geroscience: a multidisciplinary approach to understanding HIV disease

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Society is aging

- 2017: people aged >65= 17% of the population

- 2030: youngest baby boomers reach 65: elderly = ~26% of the population

- HIV: Dutch AHENA cohort
  > age 50: 28% in 2010; 73% by 2030
These demographic trends carry enormous socioeconomic and health care implications.

Aging is a relatively ignored topic in the national and international debate

Negative attitudes/stereotypes about aging

Preparing for an Aging World: Engaging Biogerontologists, Geriatricians and the Society

Recent scientific advances suggest that slowing the aging process (senescence) is now a realistic goal. Yet most medical research remains focused on combating individual diseases. Using the Future Elderly Model—a microsimulation of the future health and spending of older Americans—we compared optimistic “disease specific” scenarios with a hypothetical “delayed aging” scenario in terms of the scenarios’ impact on longevity, disability, and major entitlement program costs. Delayed aging could increase life expectancy by an additional 2.2 years, most of which would be spent in good health. The economic value of delayed aging is estimated to be $7.1 trillion over fifty years. In contrast, addressing heart disease and cancer separately would yield diminishing improvements in health and longevity by 2060—mainly due to competing risks. Delayed aging would greatly increase entitlement outlays, especially for Social Security. However, these changes could be offset by increasing the Medicare eligibility age and the normal retirement age for Social Security. Overall, greater investment in research to delay aging appears to be a highly efficient way to forestall disease, extend healthy life, and improve public health.
The Geroscience Hypothesis:

“aging is the major modifiable risk factor for most chronic diseases”

(Hodes et al. Ann NY Acad Sci Dec. 2016)

Trans-NIH Geroscience Interest Group formed in 2012
(NIA + 20 other institutes/centers)
PILLARS OF GEROSCIENCE

(Disease Drivers of Aging. Hodes et al. Ann NY Acad. Sci 2016)
A Systems Approach to Aging

LEVEL

Cell

Organ

Organism (Person)

Group (Family, Ethnic Group)

Organization (Educational Institution, Health Care)

Society (Nation)

Supranational System (Global)
Common health features of older adults

• Multiple co-morbidities
• Geriatric syndromes (e.g., falls, incontinence, sarcopenia)
• Polypharmacy
• Infection
• Social difficulties (e.g., isolation, poverty)
Aging & HIV

- Diagnosed later/treatment started later (not routinely tested, symptoms attributed to age, doctors less likely to discuss sexual activity)
- Widowed/divorced dating again; multiple partners, not aware of HIV risks
- Not concerned with pregnancy
- Stigma: do not disclose HIV status
- Double jeopardy: CVD, bone loss, cancer
- Risk of HIV drug interaction with meds associated with chronic diseases
Older HIV+ persons

- Cardiovascular disease
- Renal impairment
- Osteoporosis
- Non-AIDS-related forms of cancer
- Neurocognitive problems
- frailty

Non-AIDS-related mortality has eclipsed AIDS-related mortality as major cause of death in persons treated with antiretroviral therapy
Geroscience: a multidisciplinary approach to understanding HIV disease
B CELLS PRODUCE ANTIBODIES: the receptor is a small sample of the particular antibody that the B cell can produce.

B cells can recognize “free” antigens.
Cytotoxic T cell recognizes complex of viral peptide with MHC class I and kills infected cell

At least $10^{12}$ different types of T cell antigen receptors
MASSIVE PROLIFERATION REQUIRED
Repeated Stimulation of human T Cells

senescence
Chronic stimulation in culture of cytotoxic CD8+ T cells leads to a state of dysfunction known as **replicative (cellular) senescence**
- irreversible cell-cycle arrest
- critically short telomere length
- reduced telomerase activity
- permanent loss of CD28 surface and gene expression
- resistance to apoptosis
- altered cytokine profiles (↑pro-inflammatory)
- *Senescent CD8+ T cells correlated with reduced antibody responses to vaccines, frailty and morbidity, and chronic inflammatory diseases such as autoimmune diseases, cancer, CVD, and bone disorders*
Telomerase Counteracts this Effect

Present in germ cells, stem cells, and activated immune cells
## Effect of Repeated Stimulation on Telomerase

<table>
<thead>
<tr>
<th>Stimulation number</th>
<th>HELPER (CD4)</th>
<th>CYTOTOXIC (CD8)</th>
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<tbody>
<tr>
<td>0</td>
<td>100</td>
<td>80</td>
</tr>
<tr>
<td>1</td>
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<tr>
<td>3</td>
<td>85</td>
<td>43</td>
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<tr>
<td>7</td>
<td>75</td>
<td>10</td>
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</tbody>
</table>

Suppression of CD28 gene expression in senescent T cells

- co-stimulation
- immunological synapse
- mRNA stabilization
- trafficking
- telomerase activity
- glucose metabolism
- progressive loss of Glut1 as T cells progress to senescence in culture
CD28-negative T cells increase with age in humans (but not mice)

<table>
<thead>
<tr>
<th></th>
<th>T cells</th>
<th>in CD4</th>
<th>in CD8</th>
</tr>
</thead>
<tbody>
<tr>
<td>Neonates</td>
<td>1%</td>
<td>NT</td>
<td>NT</td>
</tr>
<tr>
<td>Young adults</td>
<td>13%</td>
<td>4%</td>
<td>42%</td>
</tr>
<tr>
<td>(20 – 40 years)</td>
<td></td>
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<tr>
<td>Elderly</td>
<td>29%</td>
<td>12%</td>
<td>69%</td>
</tr>
<tr>
<td>(70 – 90 years)</td>
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</tbody>
</table>

Boucher et al., Exp. Gerontol., 1998

Component of Immune Risk Phenotype (IRP) associated with increased mortality risk (Wikby et al., 2002; Ouyang et al., 2002)
In vivo accelerators of T cell replicative senescence

Oxidized lipids
Cortisol/stress
Chronic infection
Adenosine
Prostaglandins
LRRN3
Replicative senescence and HIV disease

- **Expansion of CD8+ T cells** during HIV disease; these cells show signs of late differentiation and senescence

- **Shortened telomeres** and low proliferative potential in **CD8+CD28- population in HIV+ persons** compared to control subjects (Effros et al. AIDS Fast Track, 1996)

- CD8+ T cells from elite controllers have significantly longer telomeres than progressors (Lichterfield et al. Blood, 2008)

- High proportion of CD8+CD28- T cells early in disease correlates with increased rate of progression to AIDS (Cao et al. J. Acquir Immune Defic Syndr 2009)

- Greying of HIV+ population: enhanced risk in terms of replicative senescence
CD28-negative T cells have short telomeres

Effros et al., AIDS 10:F17, 1996

- Unable to proliferate
- Accumulate over time
HIV induces changes in the T-cell compartment

- Telomere shortening in all three T cell subsets with age and HIV


Graph showing the absolute number of senescent cytotoxic T-cells:
- Uninfected: 0
- Infected No ART: 250
- Infected On ART: 150

Statistical significance:
- p = 0.02
- p = 0.002
Novel biomarkers of T cell senescence: 
(1) Adenosine deaminase (ADA)

- Enzyme that converts adenosine (immunosuppressive) to inosine

- ADA/CD26 complex is a component of the immunological synapse and delivers a costimulatory signal upon T cell stimulation

- ADA associated with higher telomerase activity
  • even within the CD28+ T cell population, having ADA increases telomerase activity above that seen in CD28+ T cells lacking ADA expression (Parish et al., 2010)

- HIV-1 gp120 inhibits the binding of ADA to CD26

- Exposure to adenosine in culture accelerates CD8+ T cell progression to senescence
Low ecto-ADA expression correlates with high levels of activation markers in HIV+ subjects

ADA positively correlated with CD8+ telomerase

CD8+ T cells from elite controllers have significantly higher telomerase activity than progressors (Lichterfield et al. Blood, 2008)
Novel biomarkers of T cell senescence:
(2) Leucine-Rich Repeat Neuronal 3 (LRRN3)

- brain-enriched transmembrane protein
- linked to human aging in a large genetic screen
- causal network analysis: may lie upstream of CD28 (Horvath & Effros, unpublished)
- downregulated in T cells exposed to cortisol
- expression declines in senescent CD8+ T cell cultures
- decreased expression in purified senescent CD8+ T cells from HIV+ subjects
LRRN3 mRNA expression and HIV

LRRN3 negatively associated with activation (CD38+DR+)

LRRN3 positively correlated with expression of CD28

LRRN3 positively correlated with telomerase activity
Mediator of active inflammation through the promotion of vasodilation, fever and pain, and promotes tissue influx of dendritic cells, neutrophils, macrophages, and mast cells.
Prostaglandins and HIV disease

- HIV+ persons have higher serum levels of PGE$_2$

- HIV+ persons have T cells with elevated intracellular cAMP, which is known to be increased by PGE$_2$

- COX-2 inhibitor (Celecoxib) treatment leads to decreases in CD38 density and reduction of PD-1 (marker of exhaustion) on CD8+ T cells from HIV-infected persons
Proliferative potential and IL-2 transcription is reduced in the presence of isoPGE$_2$ and PGE$_2$

PGE$_2$ and isoPGE$_2$ reduce telomerase activity and increases the production of ROS in CD8$^+$ T cells
Chronic stimulation in culture of cytotoxic CD8+ T cells leads to a state of dysfunction known as *replicative (cellular) senescence*

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Senescent T cells, inflammation, HIV
T lymphocyte regulation of osteoclastogenic cytokines

Senescent CD8+ T cells: ↑ TNFα, IL-6, RANKL
Long-term exposure to C-reactive protein (CRP) increases secretion of RANKL and TNFα.

Bone loss preceded use of ART.

Inflammation, HIV and atherosclerosis

T cell activation and senescence predict subclinical carotid artery disease in HIV-infected women. Kaplan et al. JID, 2013

Well-controlled HIV patients with low Framingham risk score have a high prevalence of subclinical carotid atherosclerosis and the main risk factors are age and inflammation. Leon et al. Eur. J. Clin. Invest., 2017
Senescence features associated with Kaposi’s sarcoma in antiretroviral treated HIV infection  

*Umemori et al. AIDS, 2013*

19 KS+ individuals vs. 47 KS-  
Undetectable plasma HIV RNA, CD4+ T cells > 300 cells/ul

- KS+ cases had significantly greater frequency of CD8+ T cells that were CD57+ (P = 0.003) and CD28- (P = 0.06)

- Telomere length of PBMC negatively correlated with proportion of CD8+CD28- T cells
EPIGENETICS, AGING & HIV

CHANGES TO DNA THAT ALTER GENE EXPRESSION WITHOUT CHANGING THE INHERITED GENETIC CODE.

METHYLATION PATTERNS CAN BE USED TO PREDICT A PERSON’S CHRONOLOGIC AGE

(S. Horvath Genome Biol. 2013)

METHYLATION PATTERNS ARE ALSO BEGINNING TO REVEAL DIFFERENCES BETWEEN CHRONOLOGIC AND BIOLOGIC AGE

HIV-1-infection Accelerates Epigenetic Aging

- Using a multivariate model that includes methylation patterns, age, and HIV status, it is estimated that HIV-1-infection accelerates aging by ~14 years.

- HIV-Induced Aging Related Epigenetic Patterns are positively associated with senescent T-cells: $r = 0.59 (<0.001)$
  
Peripheral blood contains only a minor proportion (~2%) of the total body T lymphocyte pool
Gut vs. Blood
(healthy individuals)

- gut contains significantly greater proportions of CD8$^+$ T cells that are: $CD45RA^-$ (memory), $CD28^-$, $CD45RA^-CD28^+$ (early memory), $CD45RA^-CD28^-$ (late memory), $CD25^+$, and $HLA-DR^+CD38^+$ (activated)
- Gut more senescent than blood ??
- gut T cells show greater telomerase activity $ex vivo$ compared to blood
- significantly lower levels of CD57 and PD-1 on CD45RO$^+$ memory cells
- Effect of HIV +/- ART on gut senescence??

Dock et al. PLOS ONE in press, 2017
Fibroblasts

• Cells with senescent markers increase in frequency in samples from elderly donors

• Rate of wound healing decreases with age (cells needed for this divide more slowly or not at all)
Senescent fibroblasts

- Alterations in function
  increased collagenase production
  (destroys collagen)

- Enhance growth of tumor cells
  creates favorable microenvironment
Early passage ("young") fibroblasts

Senescent fibroblasts

Tumor cells

Replicative Senescence Prevention / Remediation

• Prophylactic vaccination against CMV, HIV-1

• Reversal of apoptosis resistance

• Modulation of replicative senescence
  
  *telomerase, CD28, TNFα*

• Physical removal of senescent T cells

Effros et al., Immunol. Revs., 2005
Removal of senescent cells

Killed off senescent cells

Kidneys, hearts: younger
Lived 20-30% longer

To stay young, kill zombie cells (Nature, 25 October, 2017)

“I lose sleep at night because these things always look good in mice or rats, but when you get to people you hit a brick wall,” says James Kirkland
Summary

• Chronic stimulation of human T cells in culture leads to a terminal state known as replicative senescence

• CD8 T cells with a similar phenotype accumulate in vivo during aging and chronic HIV-1 infection (and certain cancers)

• Driving force: latent infection (CMV, HIV-1), oxidative, mental stress, CRP.

• Senescent T cells contribute to the pro-inflammatory milieu: may affect skeletal and cardiovascular systems, and tumor microenvironment

• Reduction in the proportion of senescent T cells may impact multiple age-related pathologies

• Multiple disciplines required to elucidate the complex relationship between chronological aging and chronic HIV infection
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