Buck Institute for Research on Aging

16 February, 2021

NIH DNA repair interest group

DNA damage and repair: Balancing cancer and aging

Lawrence Berkeley National Laboratory

Disclosure:

I am a scientific co-founder of UNITY Biotechnology

Aging = susceptibility to (chronic) disease

not a coincidence! caused by basic aging process(es)

Neurodegeneration, memory loss

Osteoporosis

Macular degeneration, hearing loss

Heart disease

Vascular disease

Sarcopenia, frailty

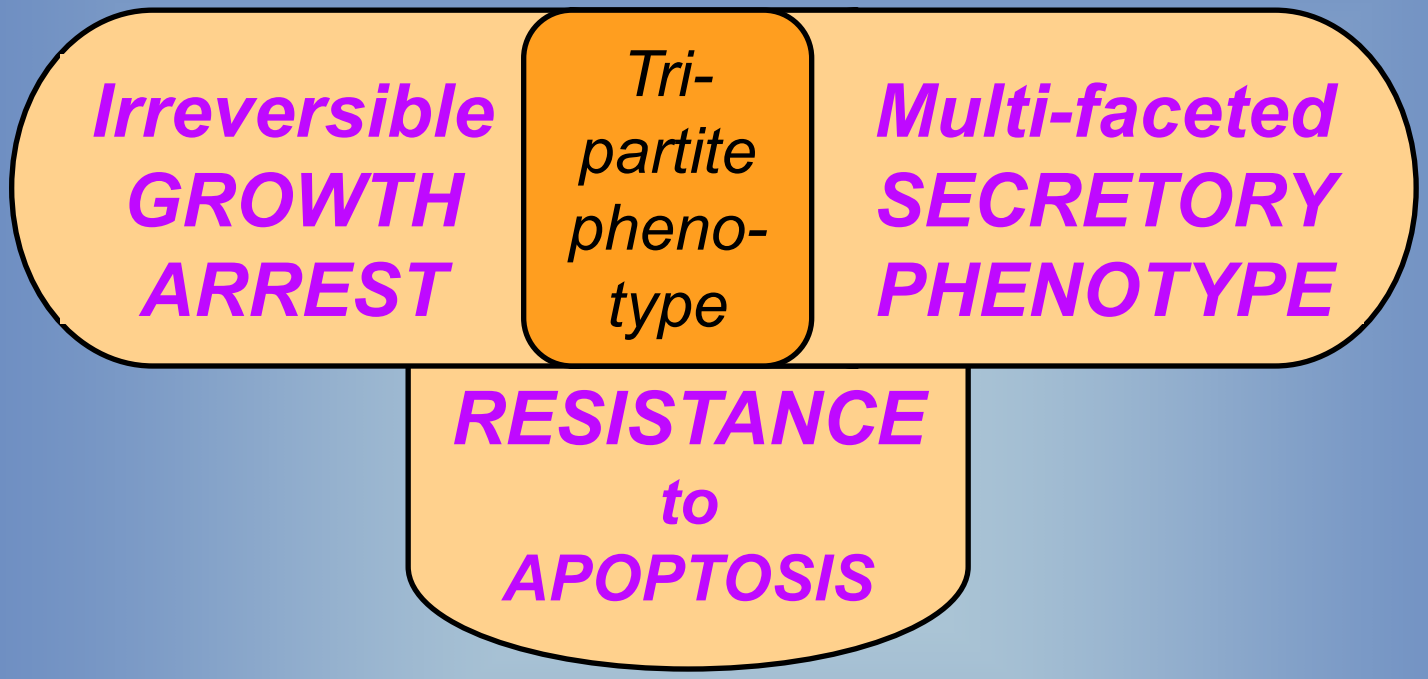
Diabetes, metabolic syndrome

Decreased lung, kidney, etc function

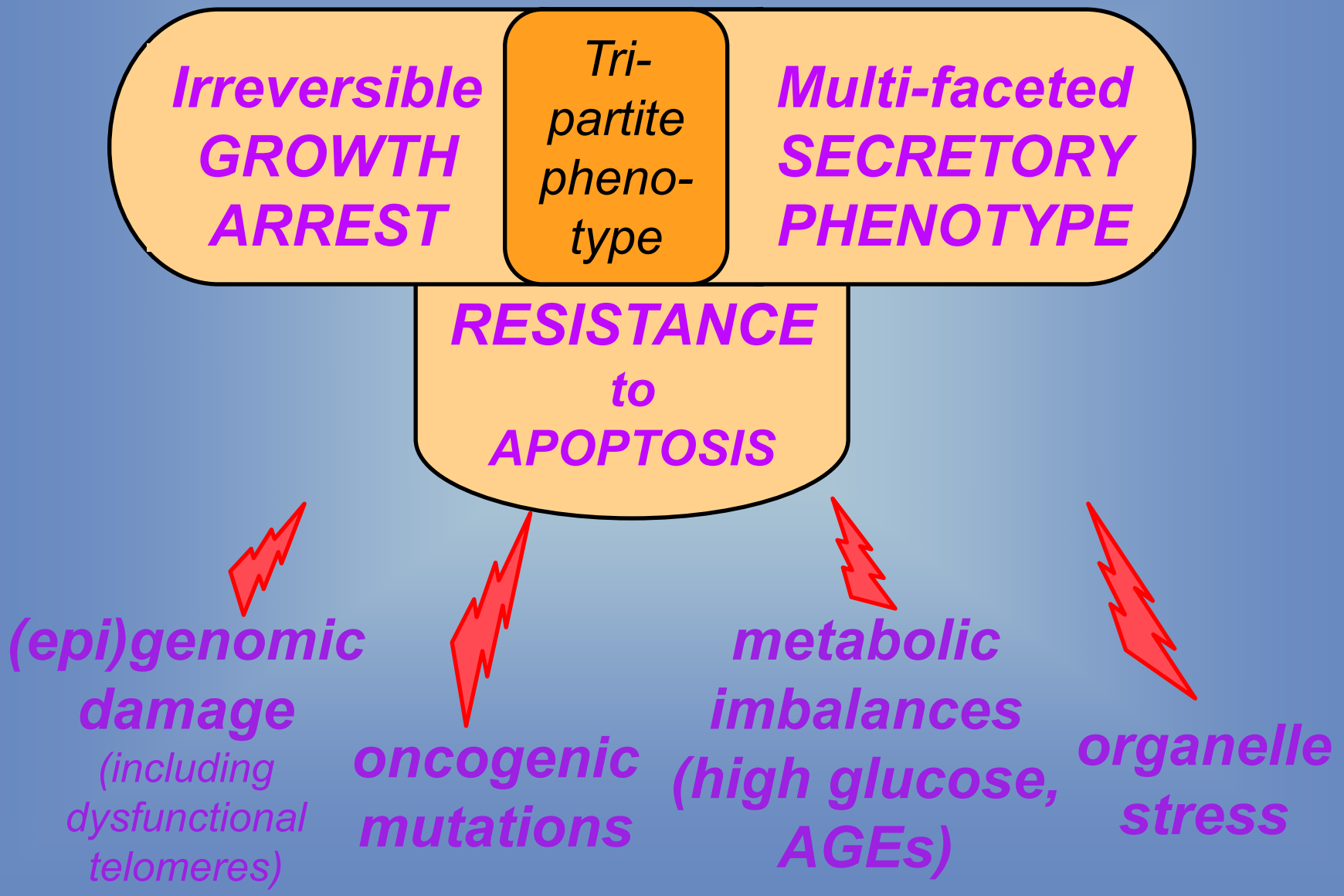
CANCER

Cellular senescence: a candidate basic aging process

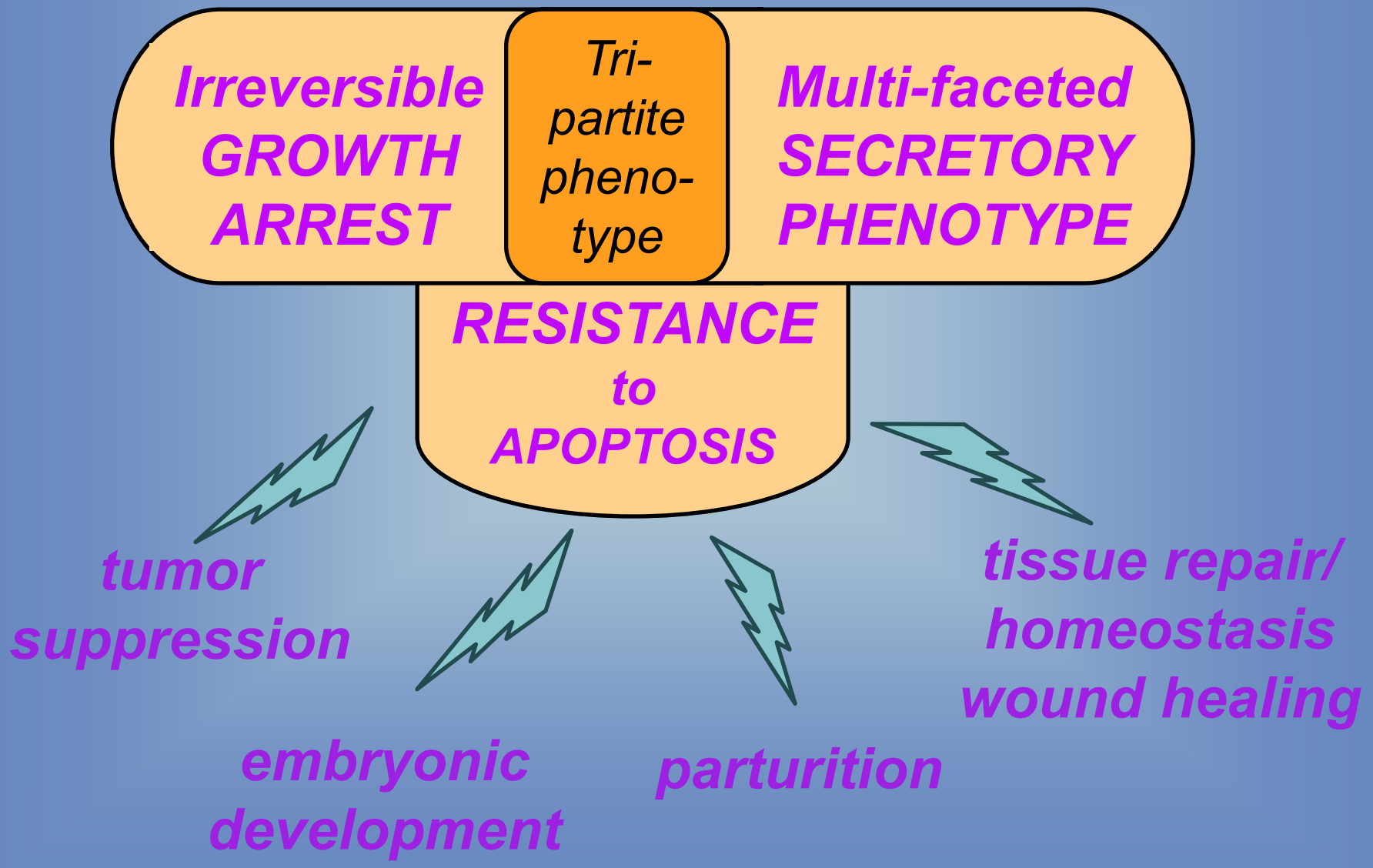
What is cellular senescence?



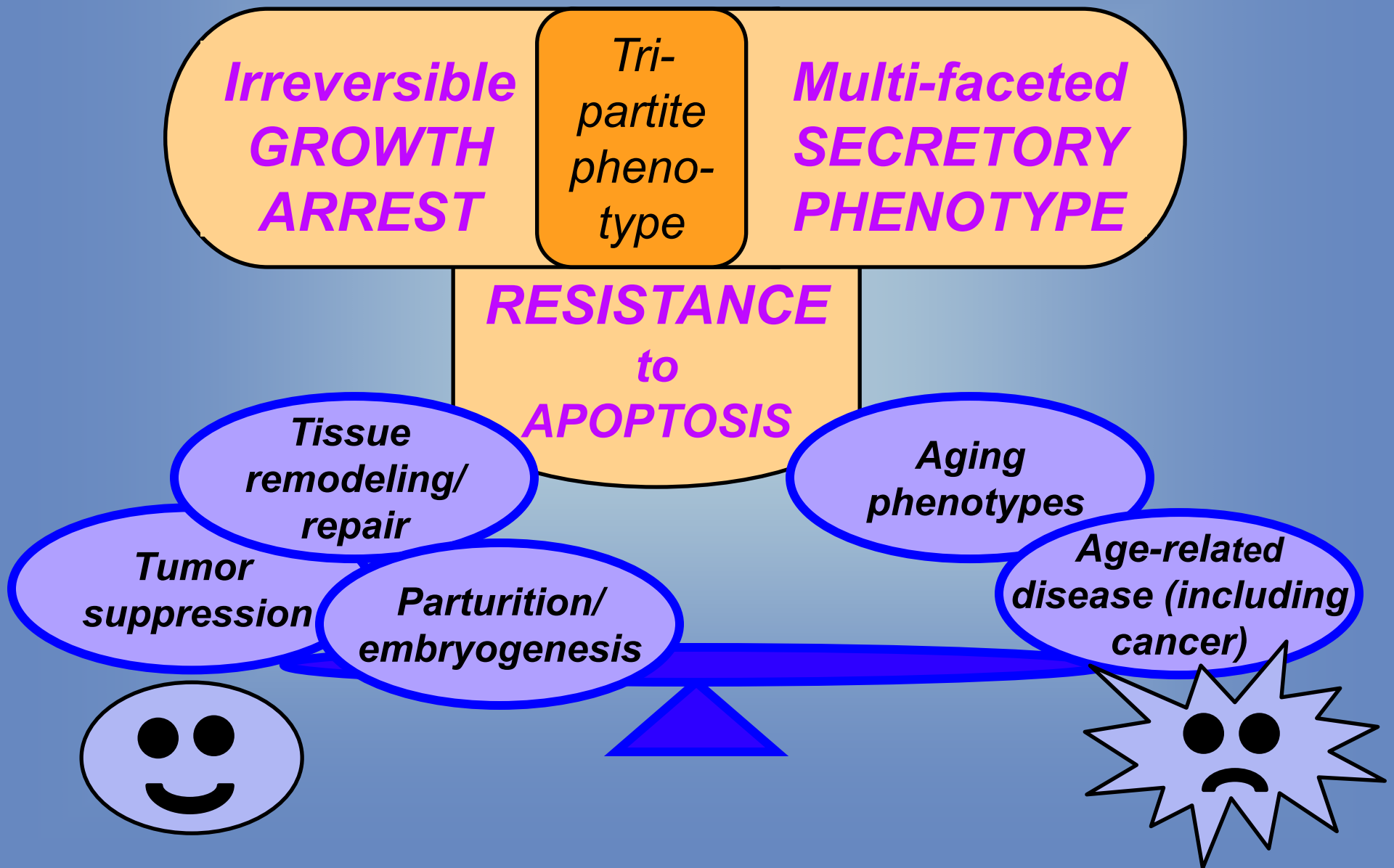
Cellular senescence, a complex response to stress (many cancer-causing)



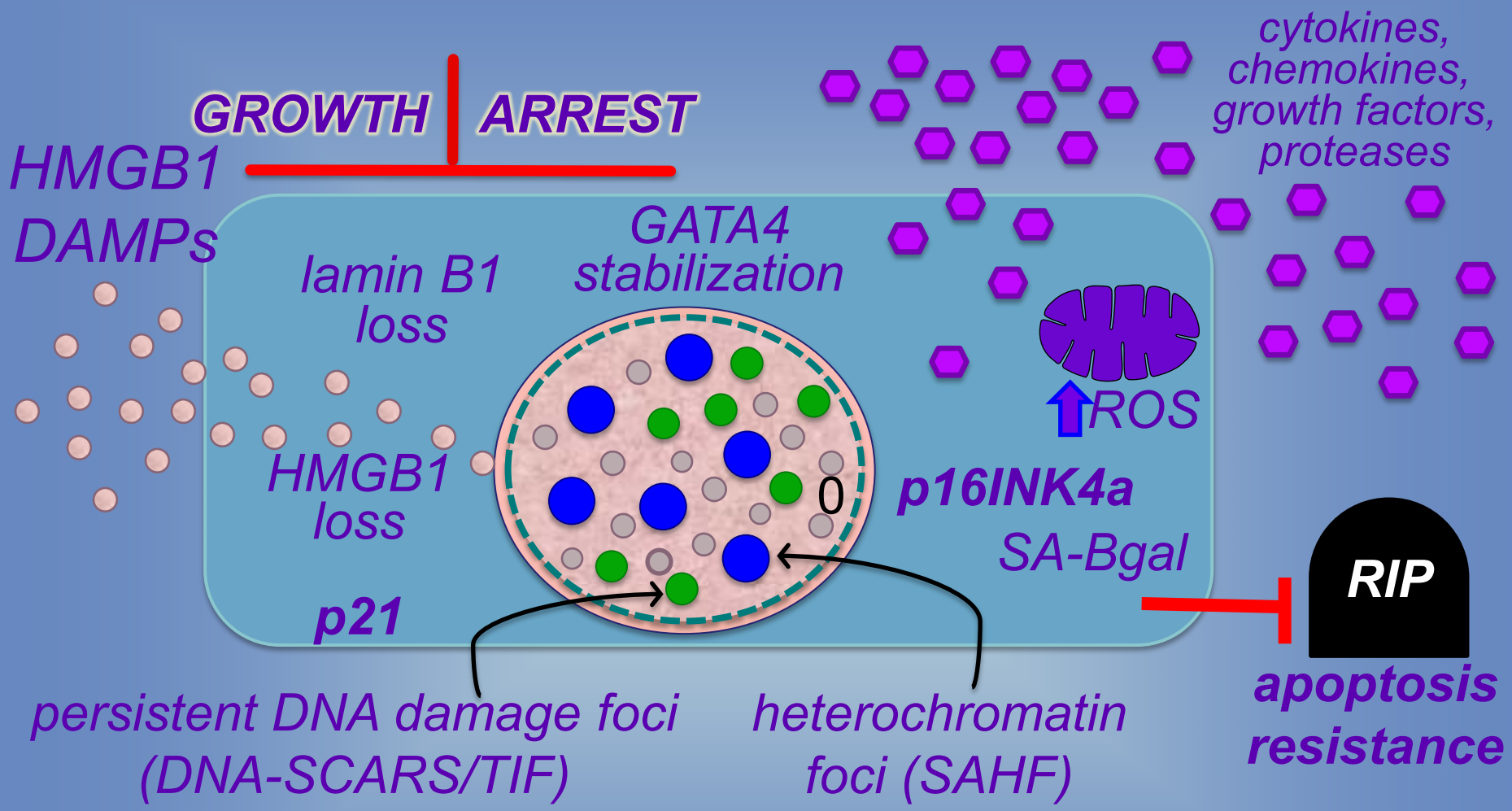
Cellular senescence, a physiological response (promotes health)



Cellular senescence, an evolutionary balancing act



What defines a senescent cell?



ps …There are no senescence-SPECIFIC markers!

Dimri et al, PNAS, 1995; Beausejour et al, EMBO J, 2003; Narita et al, Cell, 2003; Rodier et al, Nature Cell Biol, 2009; Rodier et al, J Cell Sci, 2011; Freund et al, Molec Biol Cell, 2012; Davalos et al, J Cell Biol, 2013; Kang et al, Science, 2015

When/where are senescent cells found in vivo?

With Increasing Age

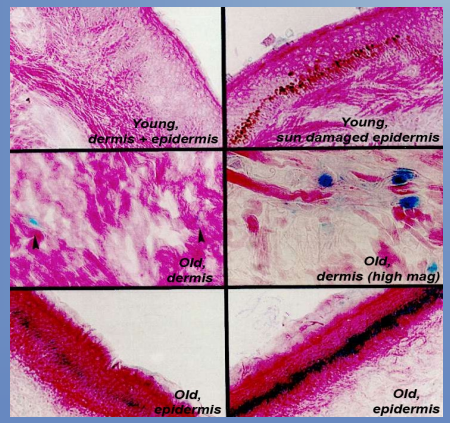
Human, non-human primates, rodents, zebrafish
skin, retina, liver, spleen, aorta, kidney, lung, etc.

At Sites of Age-Related Pathology

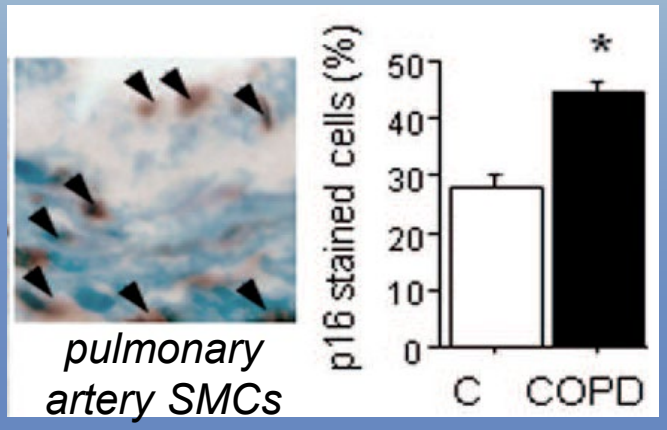
Venous ulcers, atherosclerotic plaques, arthritic joints,
COPD, visceral fat, AD brain, etc
Benign prostatic hyperplasia, pre-neoplastic lesions

Senescent cells are not abundant!

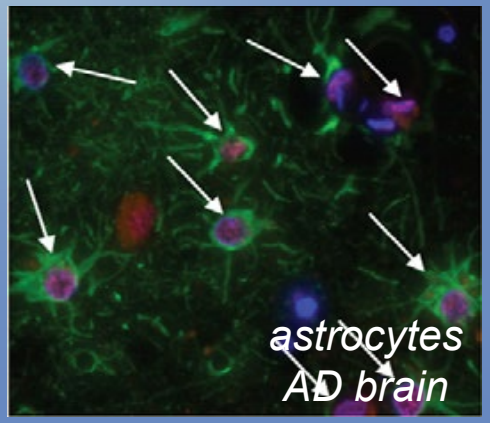
Dimri et al., PNAS, 1995



Noureddine et al., Circulation, 2011



Bhat et al, PLoS One 7, 2012

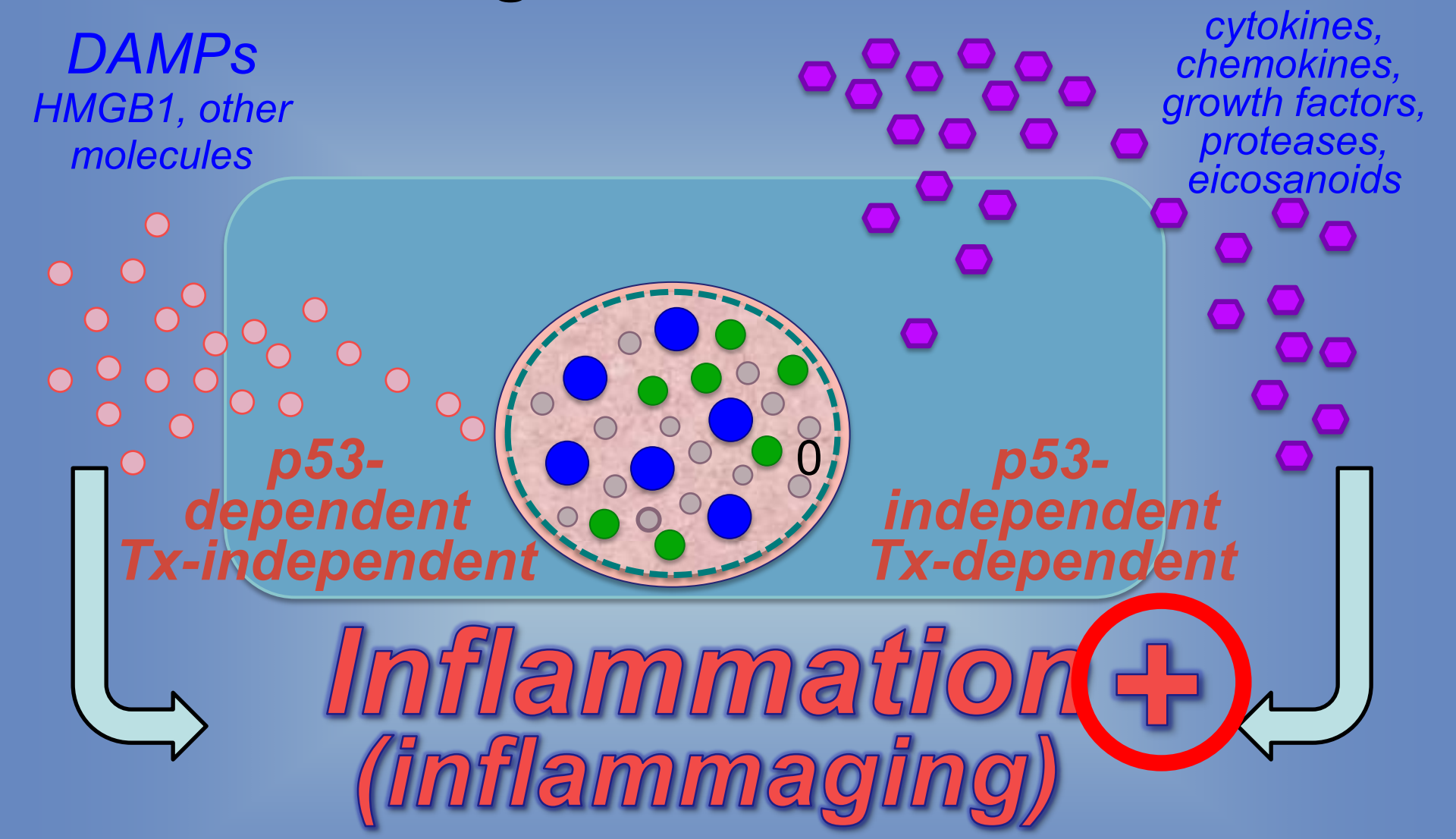


Senescent cells …. a smoking gun?

Present at the right time and place to drive aging and age-related disease

***HOW** could they drive aging phenotypes/pathology?*
*And **DO** they drive aging phenotypes/pathology?*

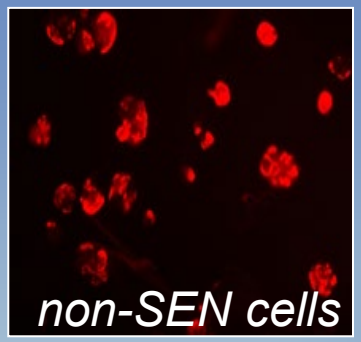
How might the SASP act?

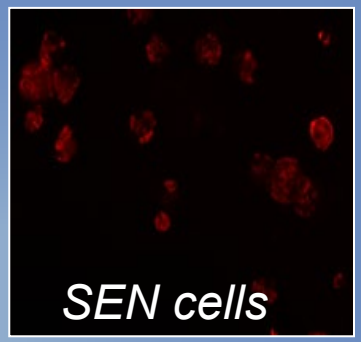


destroy tissues | **disrupt normal cell functions** | **prevent stem cell functions** | **promote cancer** | **promote tissue repair**

DO SnCs and the SASP drive aging/disease?

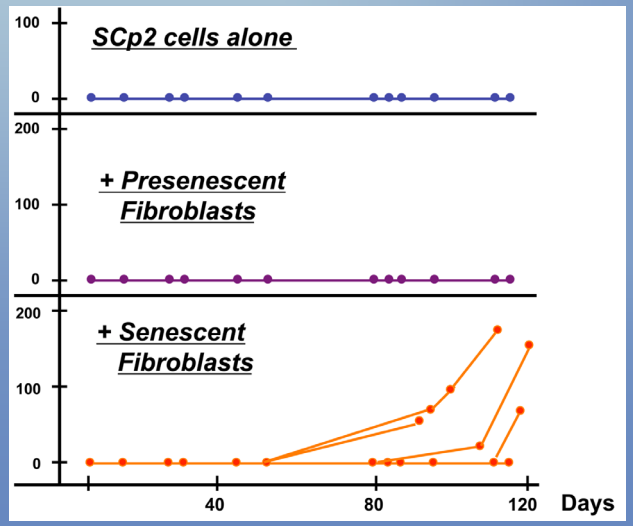
disrupts normal tissue structures and functions
(mostly due to MMPs)
Parrinello et al, 2005, J Cell Sci





disrupts normal stem cell functions
(stimulates cancer stem cell proliferation)
unpublished

promotes malignant phenotypes
Krtolica et al, 2001, PNAS
Coppe et al, 2006, J Biol Chem



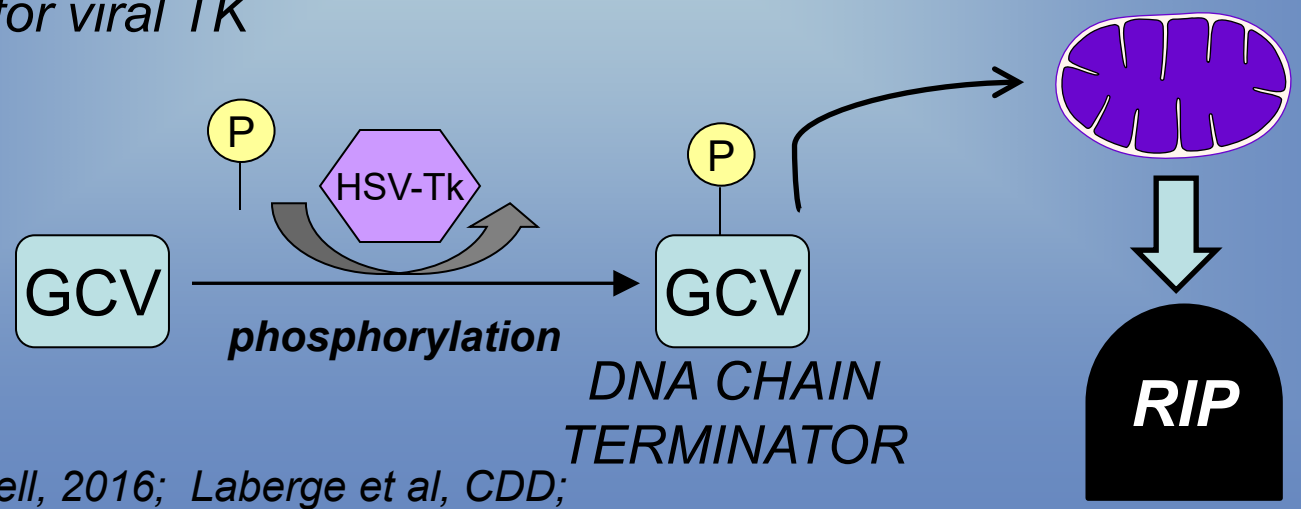
DO SnCs and the SASP drive aging/disease?

p16-3MR (tri-modal reporter) mice

BAC containing murine INK4a locus inserted into mouse genome
3MR knock-in: downstream of p16^{INK4a} promoter + inactivation of p14ARF
Mice have normal (diploid) copies of p16 and p14 genes

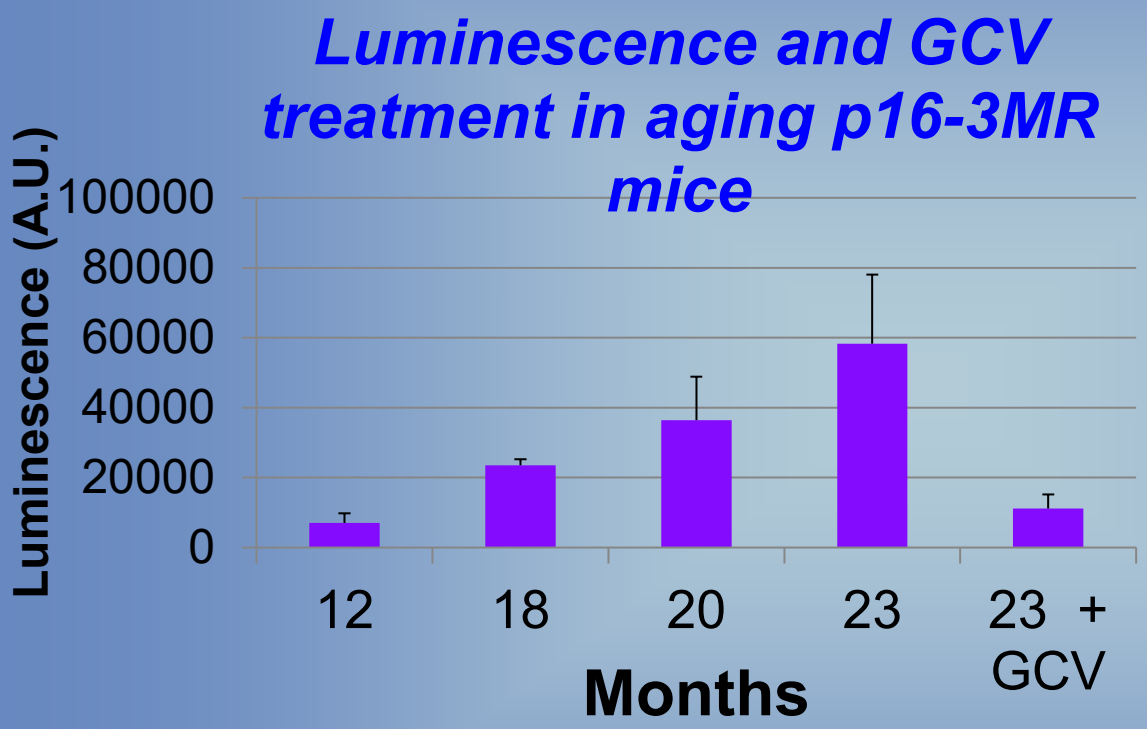
3MR: renilla luciferase; modified red fluorescent protein; herpes simplex virus thymidine kinase

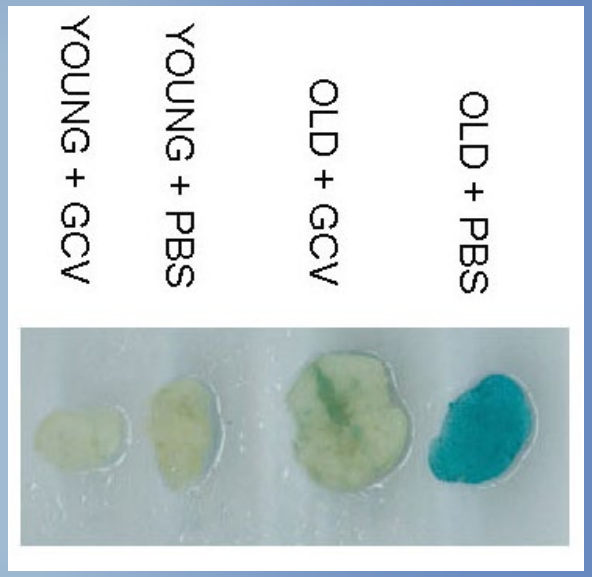
GCV = gancyclovir
Low affinity for cellular TK
High affinity for viral TK



Demaria et al, Dev Cell, 2016; Laberge et al, CDD; collaboration with Hoeijmakers, Vijg, Hasty labs

Senescent cells can be eliminated from naturally aged mice





parallel age-related increase in endogenous p16INK4a, 3MR, IL-6, etc; all reduced by GCV treatment

Demaria et al, Dev Cell, 2014

Senescent cells cause or contribute to:

Alzheimer's@@ and Parkinson's disease*
*Atherosclerosis***
*Cardiovascular dysfunction**#*
*Cancer metastasis and recurrence****
*Chemotherapy (HAART) cardiotoxicity, blood clots, fatigue****
Cognitive decline/loss of neurogenesis
*Cataracts**#*
Diabetes and diabetic complications@
Metabolic syndrome@@
Myeloid→lymphoid skewing #
*Pulmonary fibrosis#**
Osteoarthritis ##
Osteoporosis ###
Sarcopenia/frailty@@@

Wound healing, tissue regeneration@
*Embryogenesis^, parturition^^*

**Chinta et al, Cell Reports, 2018; **Childs et al, Science, 2016; **#Baker et al, Nature, 2016; ***Demaria et al, 2017; #Chang et al, Nature Med, 2016; #*Schafer et al, Nature Comm, 2017; ##Jeon et al, Nature Med, 2017; ###Farr et al, Nature Med, 2017; @ Demaria et al, Dev Cell, 2014; @Krizhanovsky et al, Cell, 2008; @@Bussian et al, Nature, 2018; @@Aguayo-Mazzucato et al, Cell Metab, 2019; @@Palmer et al, Aging Cell, 2019; @@Zhu et al, PLoS One, 2019; ^Munoz-Espin, Cell, 2013; ^Storer, Cell, 2013; ^^Menon, Aging, 2016*

Are there pharmacological agents that mimic the transgenes?

Senolytics – a new class of drugs that 'selectively' kill senescent cells

Senomorphics – drugs that 'selectively' suppress certain SASP modules

Why more basic research is needed before senolytics or senomorphics are ready for clinical use:

No known senolytic/senomorphic is efficacious against ALL SnCs

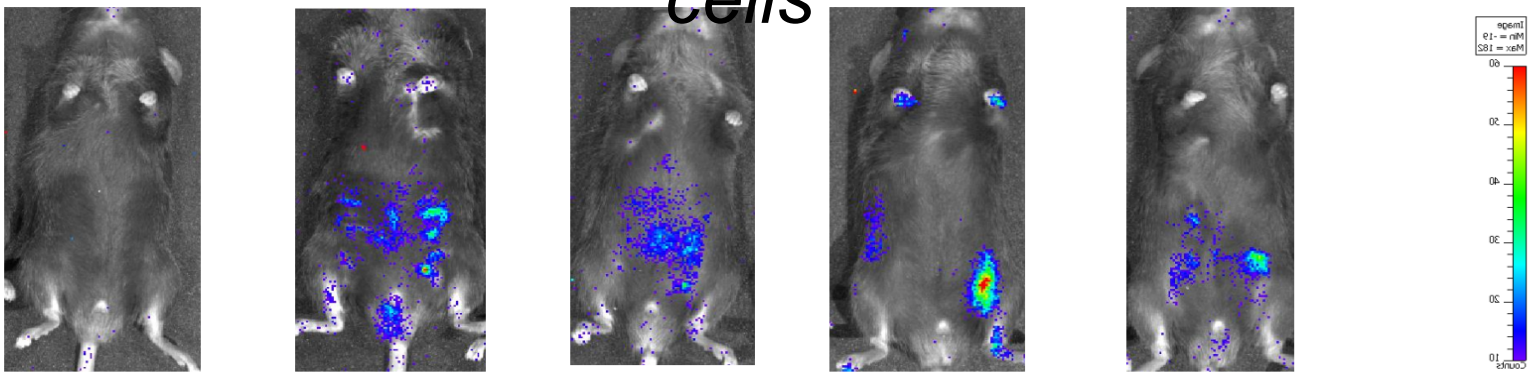
Some SnCs are beneficial (at least transiently)

The SASP is remarkably plastic

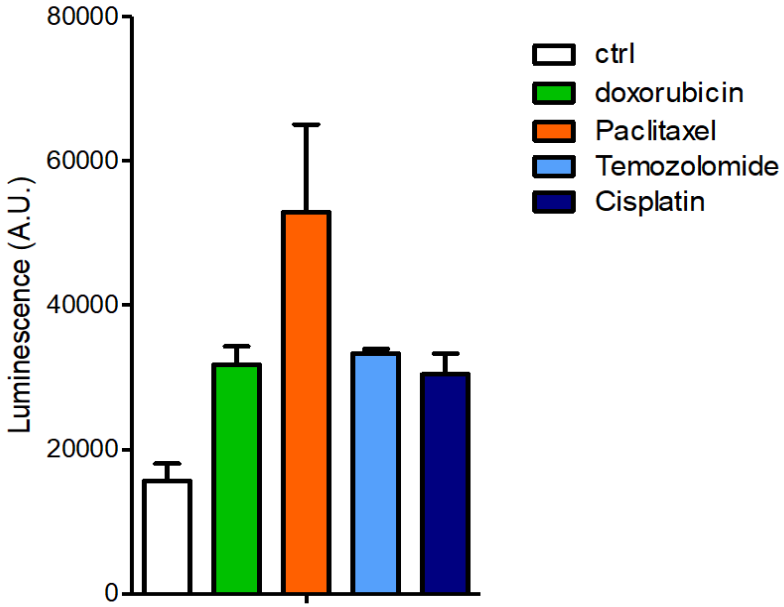
Cellular senescence and 'accelerated' aging

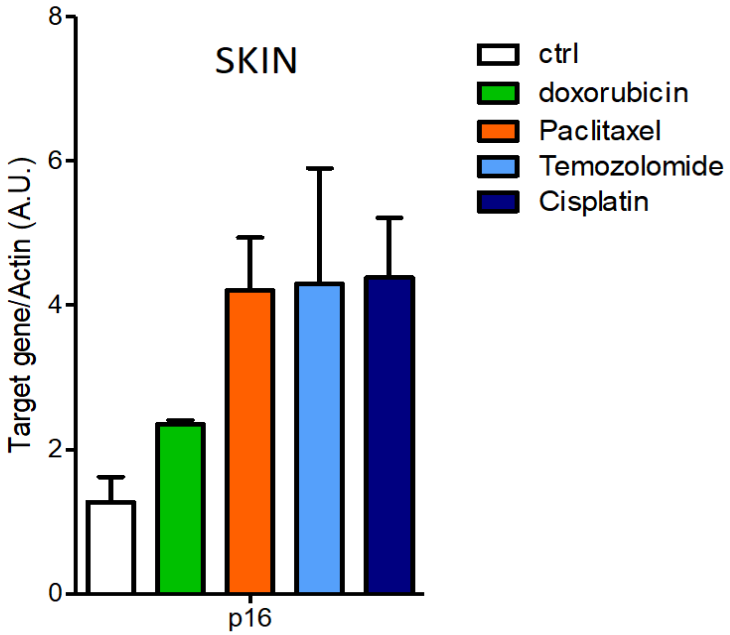
Adverse effects of genotoxic chemotherapies (and HAART therapies)

DNA damaging therapies → persistent senescent cells



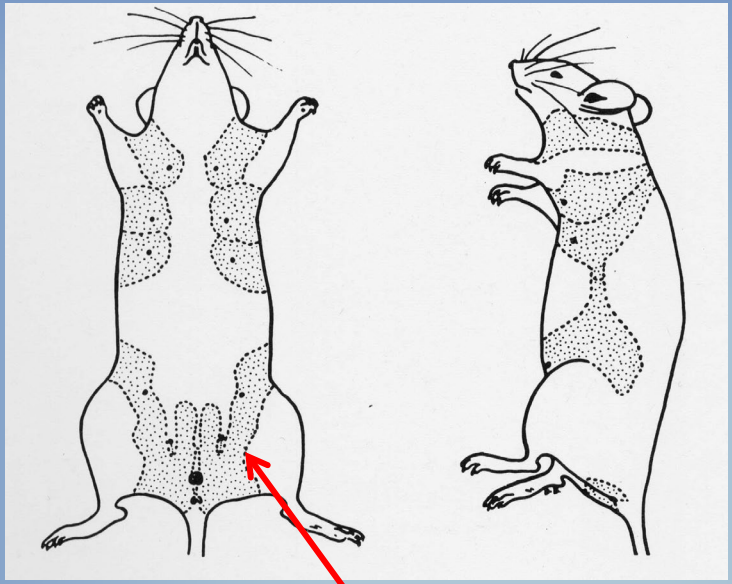
ctrl doxorubicin paclitaxel cisplatin temozolomide

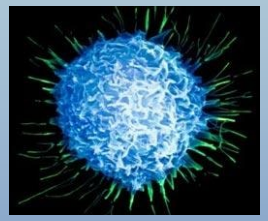




Demaria et al, Canc Disc, 2017

Senescent cells promote metastases MMTV-PyMT breast cancer

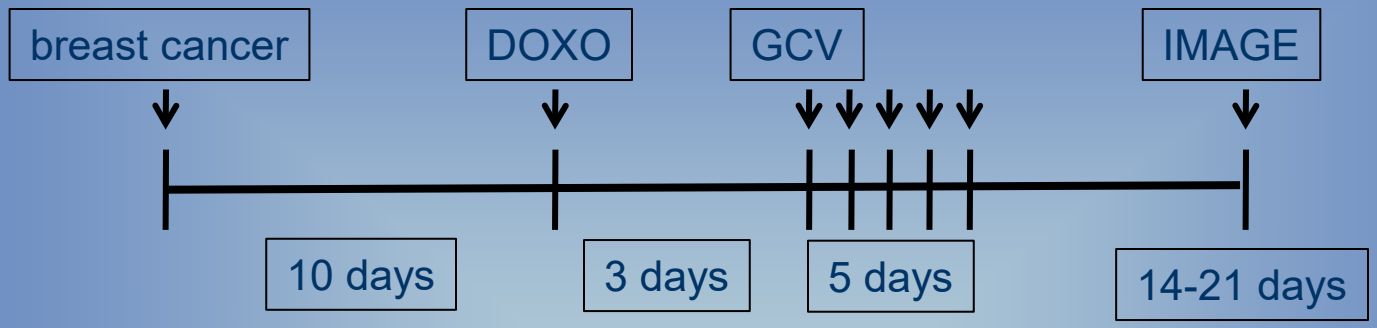




MMTV-PyMT cells expressing firefly luciferase

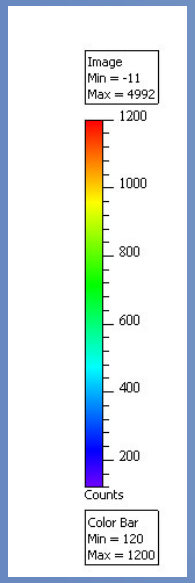
Inject into inguinal mammary fat pad → multifocal mammary adenocarcinomas + lung/liver metastases

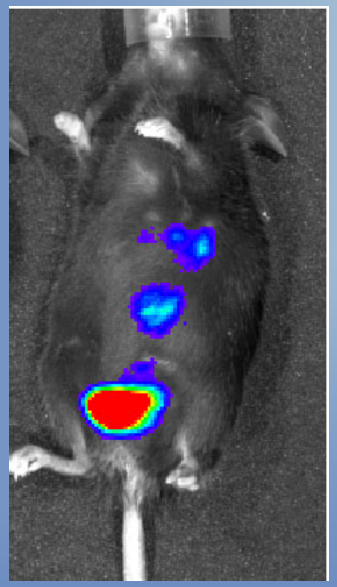
Senescent cells promote metastases in mice with breast cancer

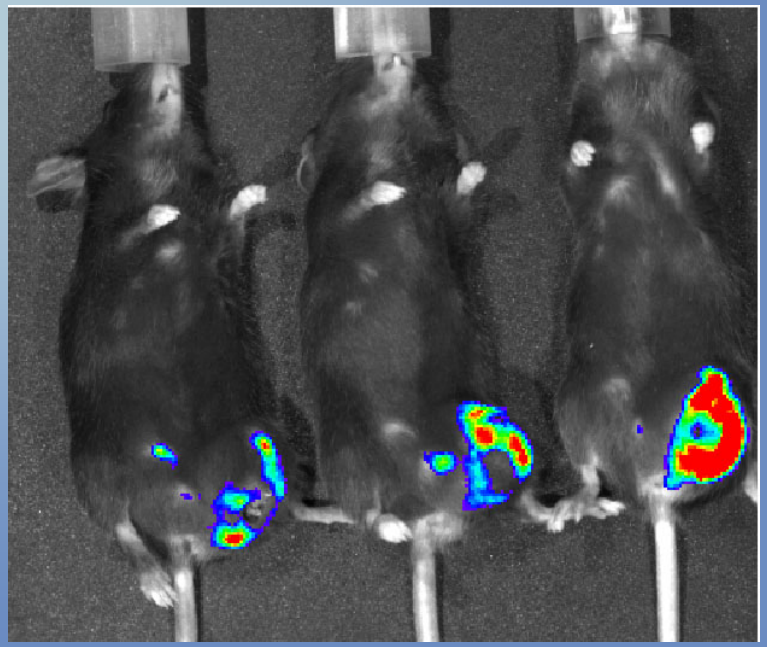


Control

GCV

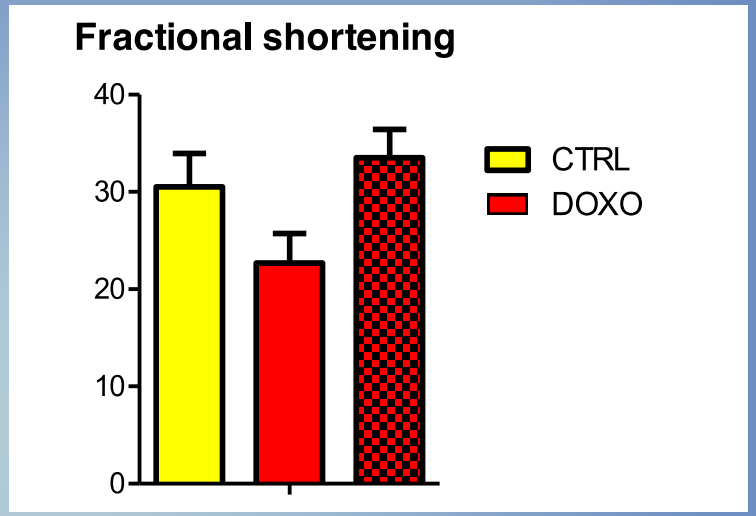






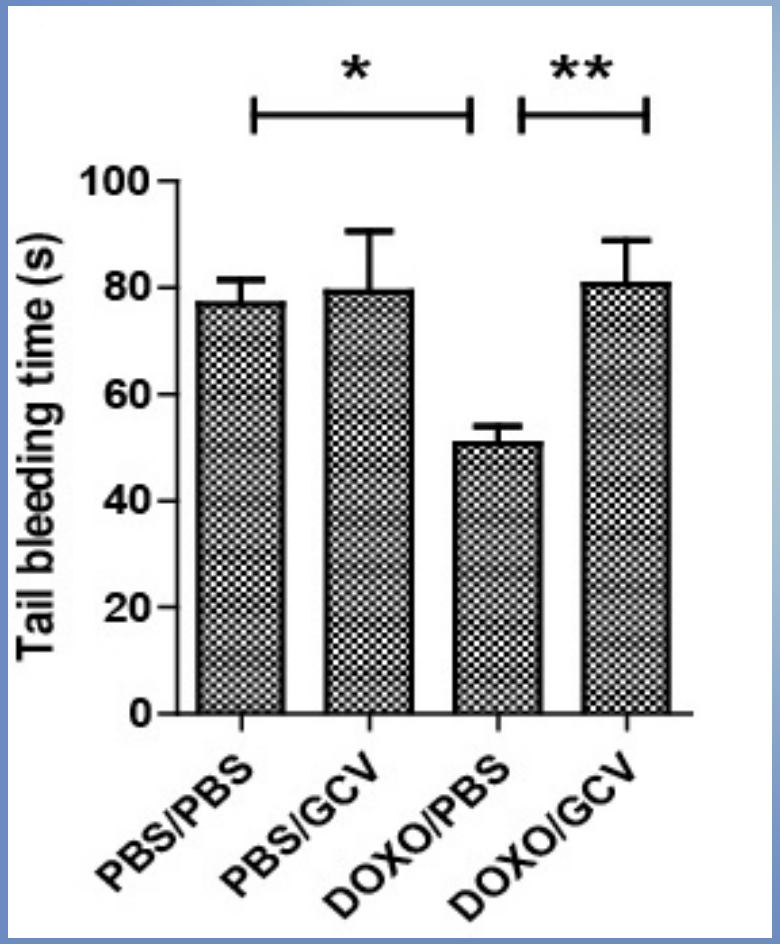
Demaria et al, Canc Disc, 2017

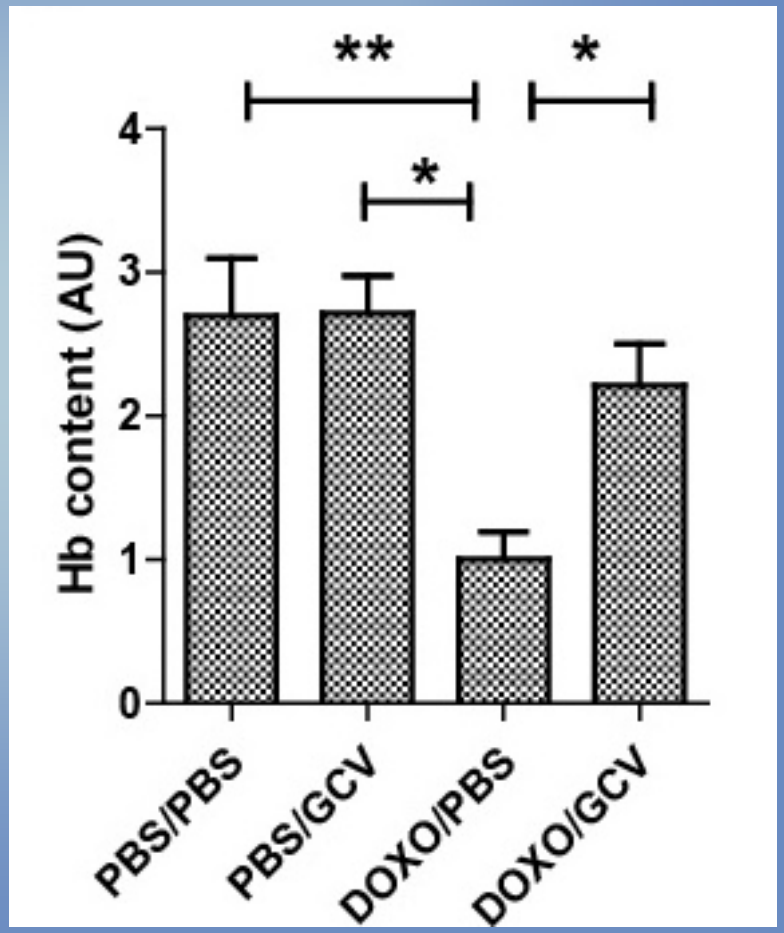
Senescent cells contribute to chemotherapy-induced cardiotoxicity



Demaria et al, Canc Disc, 2017

Senescent cells promote blood clotting





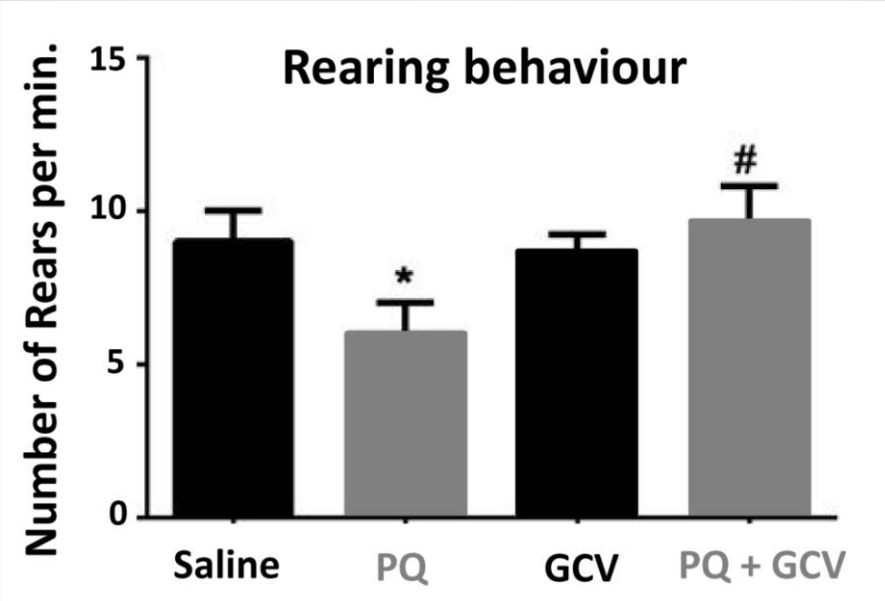
Liu et al., Cell Reports, 2019

Parkinson’s disease

Paraquat (PQ) causes oxidative damage and Parkinson's disease in mice and humans

PQ causes astrocytes to undergo senescence

PQ reduces motor neuron function; restored by GCV



Cell autonomous and non-autonomous effects:

Astrocyte-neuron interactions

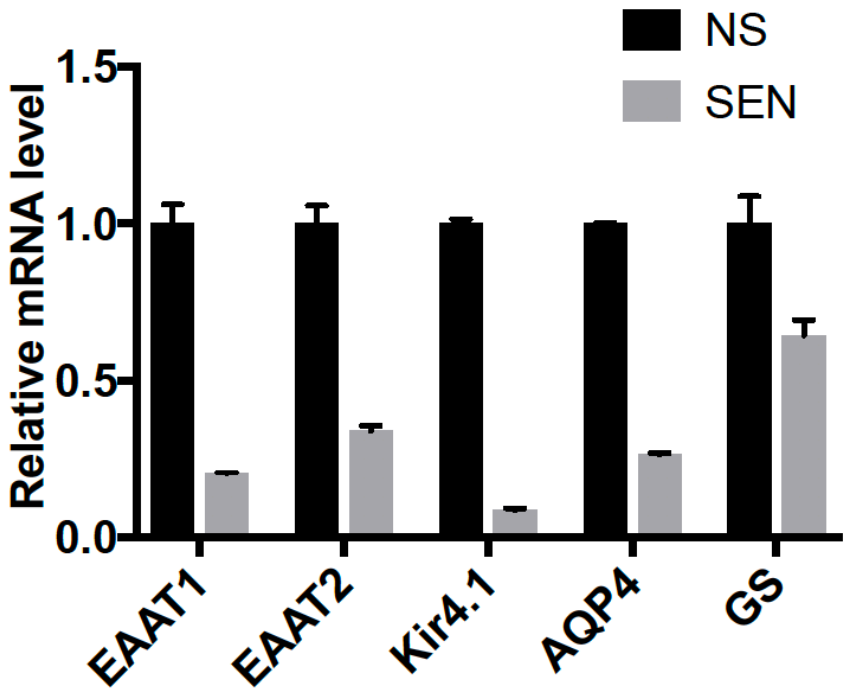
Senescent human fibroblasts and astrocyte transcriptomes (bulk RNAseq)

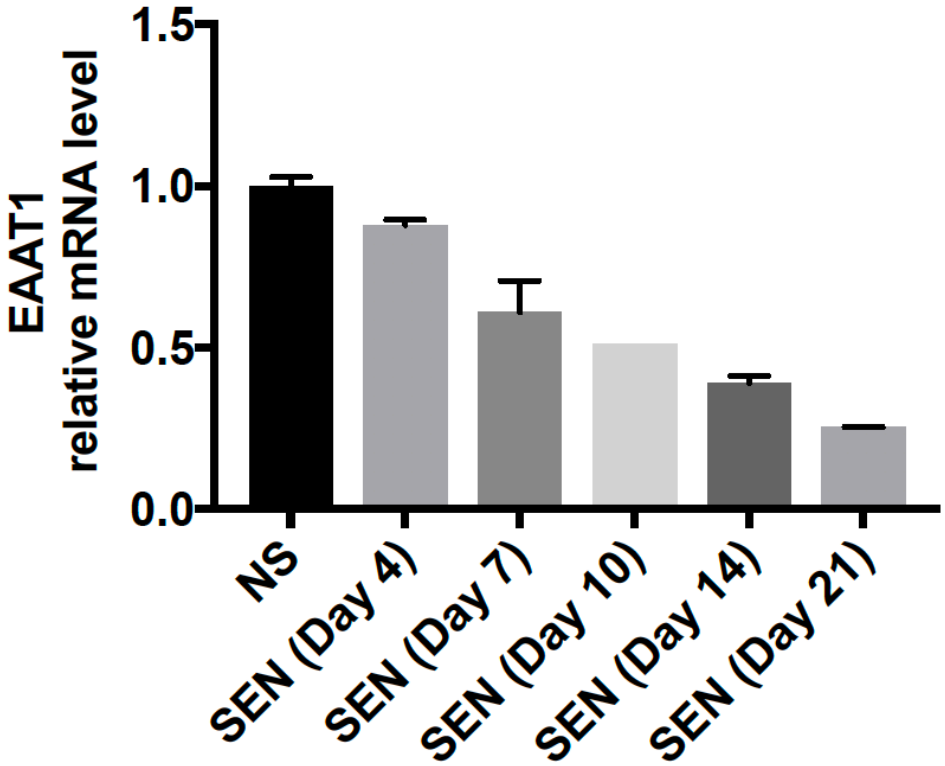
FIBROBLASTS: 70-80% mRNA changes are upregulated

ASTROCYTES: >50% mRNA changes are downregulated

Limbad et al, 2020, PLoS One

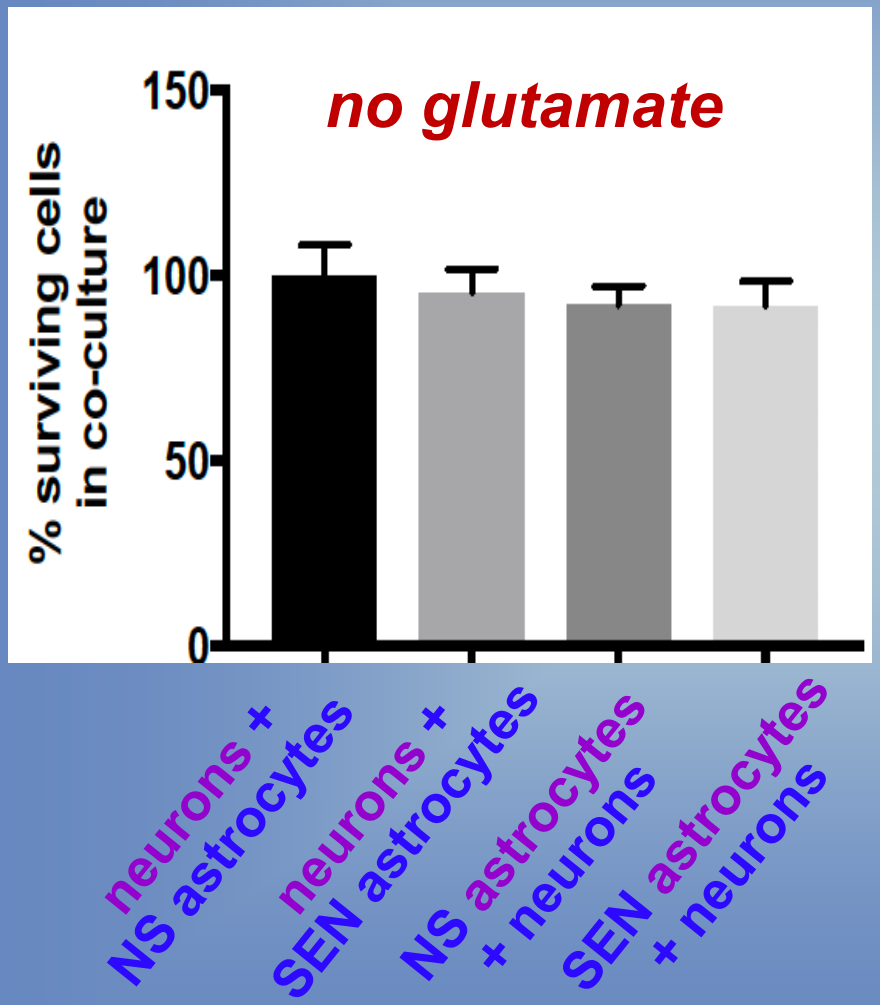
Downregulated mRNAs in senescent astrocytes encode proteins important for glutamate uptake and metabolism

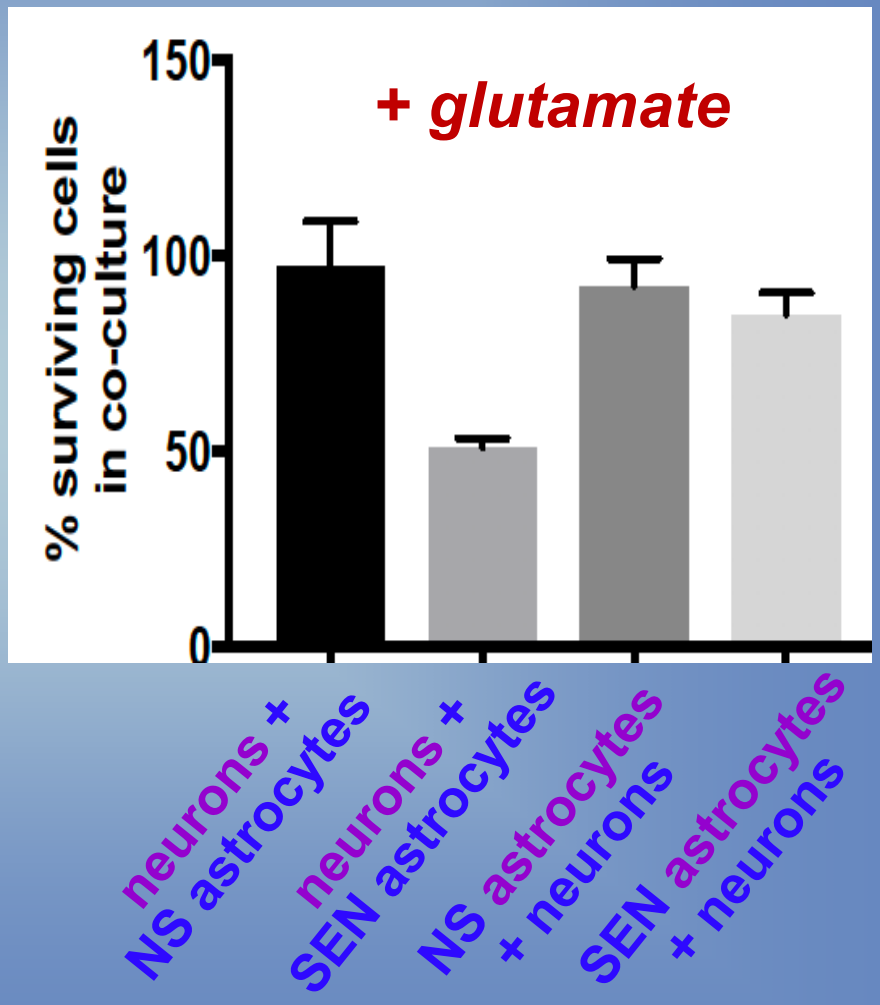




Limbad et al, 2020, PLoS One

Senescent astrocytes confer glutamate sensitivity on neurons





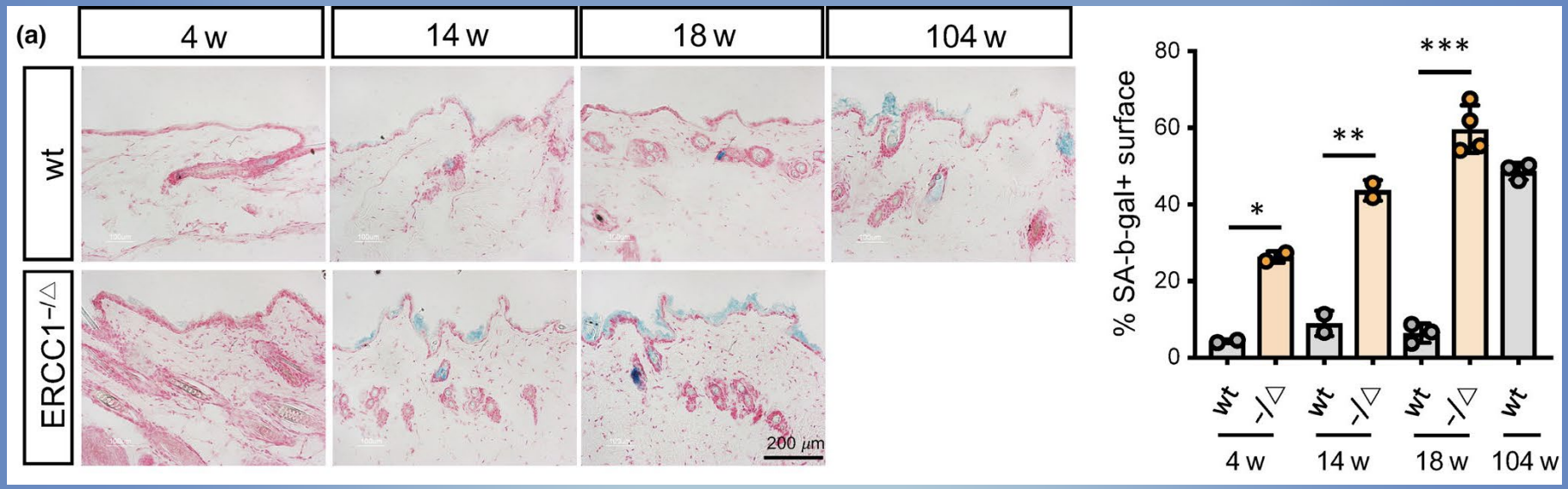
Limbad et al, 2020, PLoS One

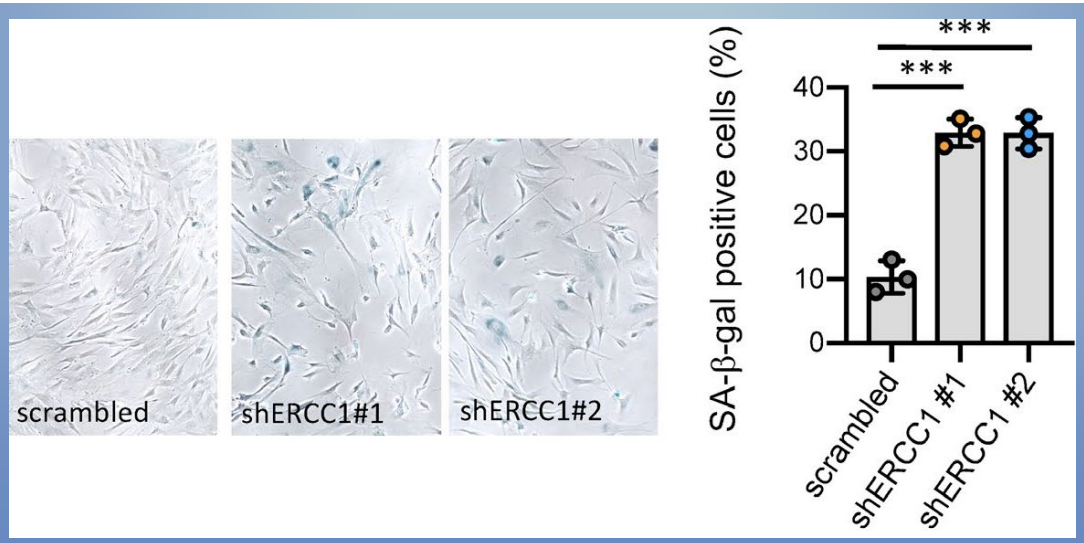
Cell autonomous and non-autonomous effects:

Effects of transcription-blocking DNA lesions

DNA damage deficient mice: ercc1-/△

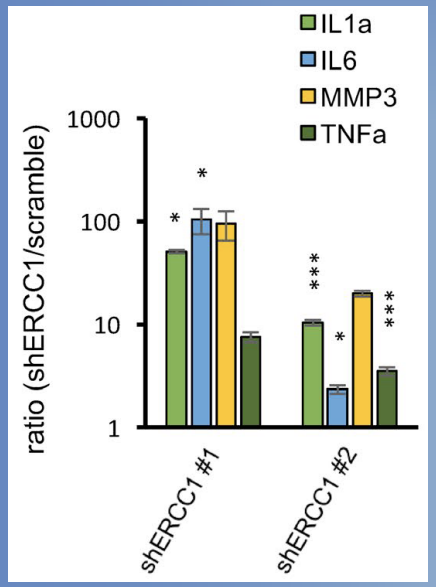
→ transcription blocking lesions (TBLs)

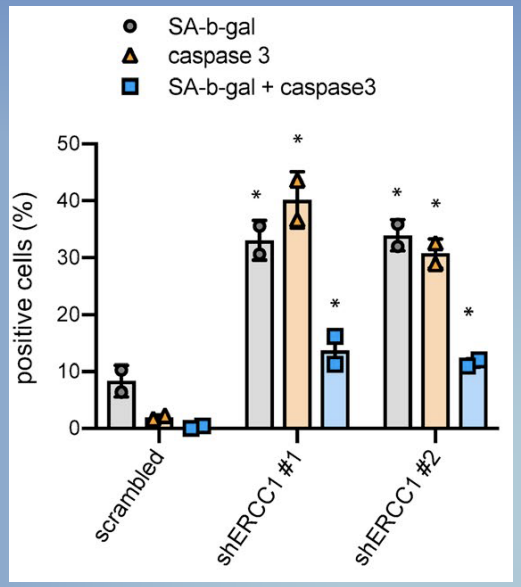


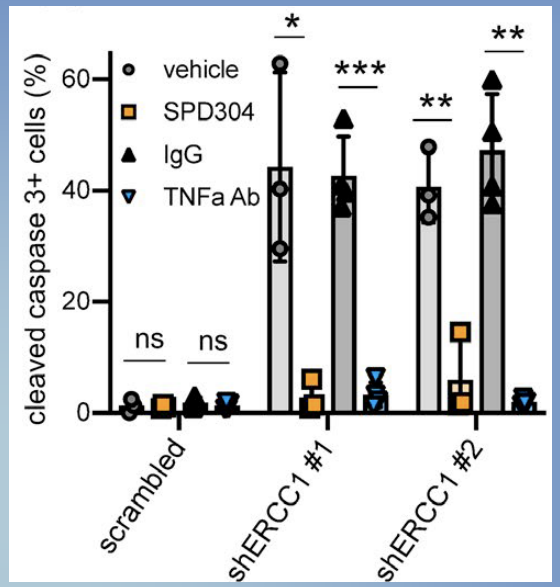


Kim et al, 2020, Aging Cell

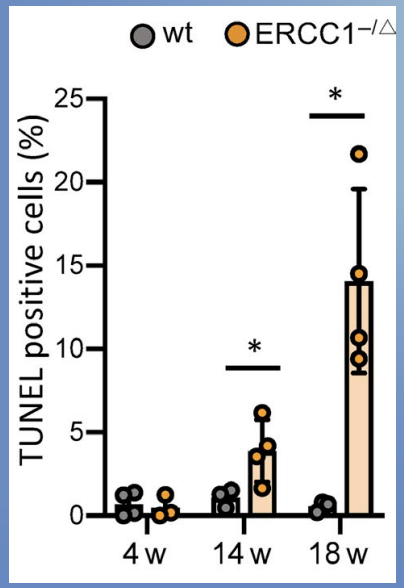
TBLs → TNFa → cell death

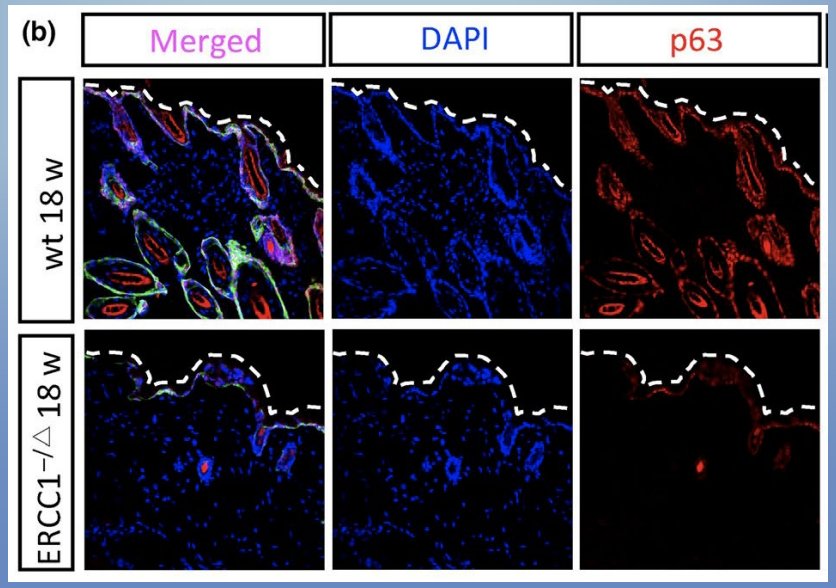






human cells





mouse skin

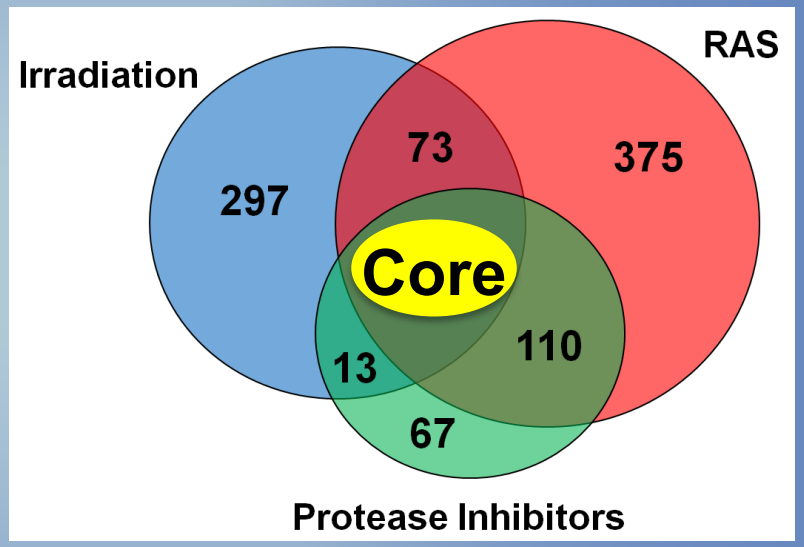
The SASP is highly complex

	Soluble SASP	Exosome SASP
X-ray irradiation	1505 proteins ↑ 554 (531 > 2-fold increase) ↓ 38 (32 > 2-fold decrease)	1502 proteins ↑ (111 > 2-fold increase) ↓ (221 > 2-fold decrease)
Oncogenic RAS	1693 proteins ↑ (675 > 2-fold increase) ↓ (10 > 2-fold decrease)	354 proteins ↑ 22 (19 > 2-fold increase) ↓ 20 (14 > 2-fold decrease)
Atazanavir	441 proteins ↑ 62 (271 > 2-fold increase) ↓ 14 (7 > 2-fold decrease)	

966 soluble SASP factors
470 exosome SASP factors

SASP Atlas (Buck web site)

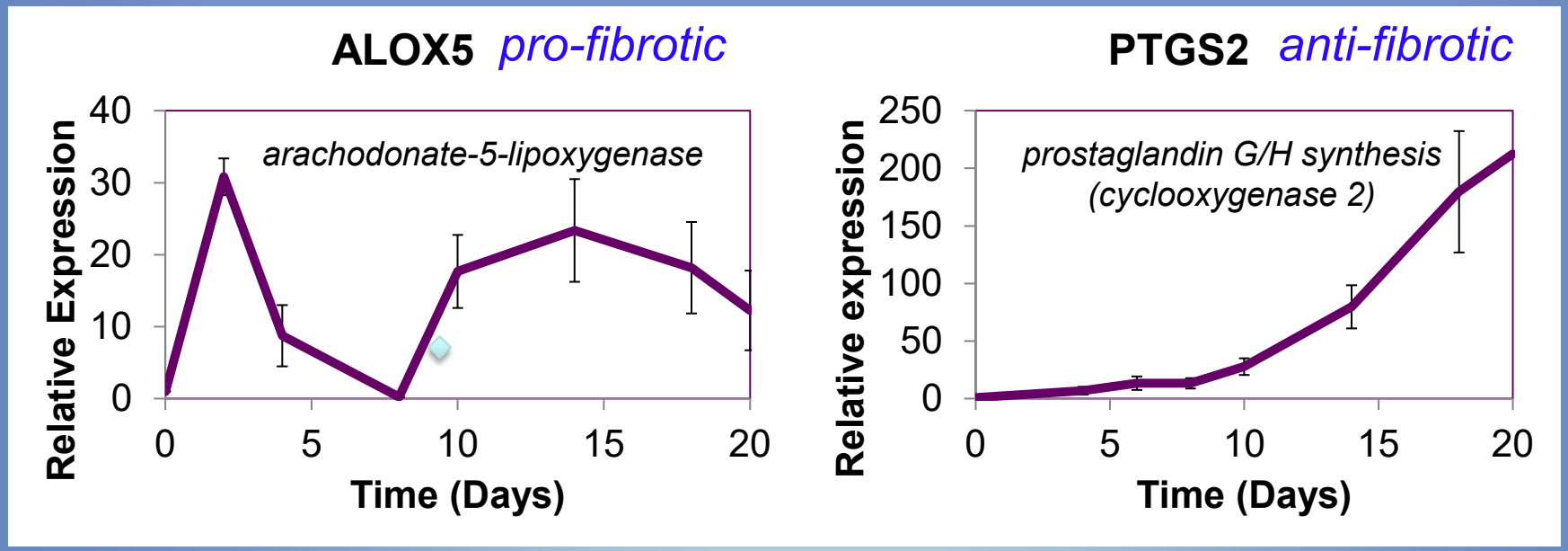
Core SASP proteins

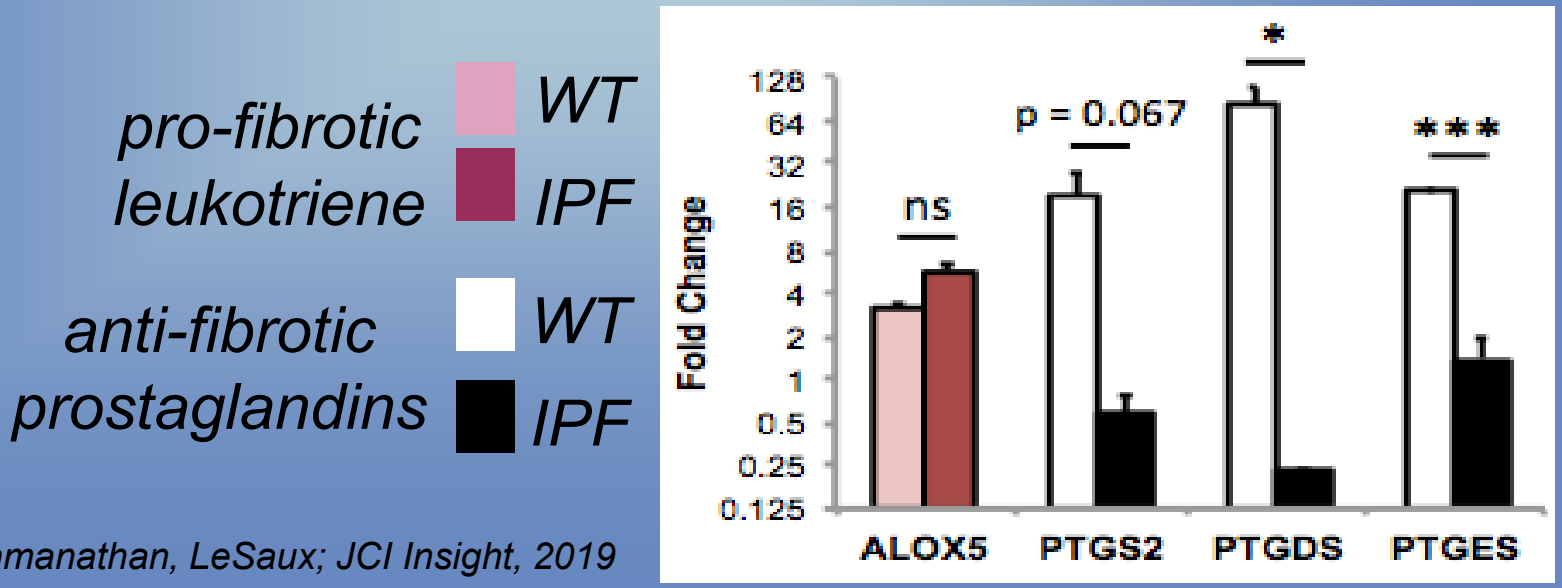


+ dozens of bioactive lipids

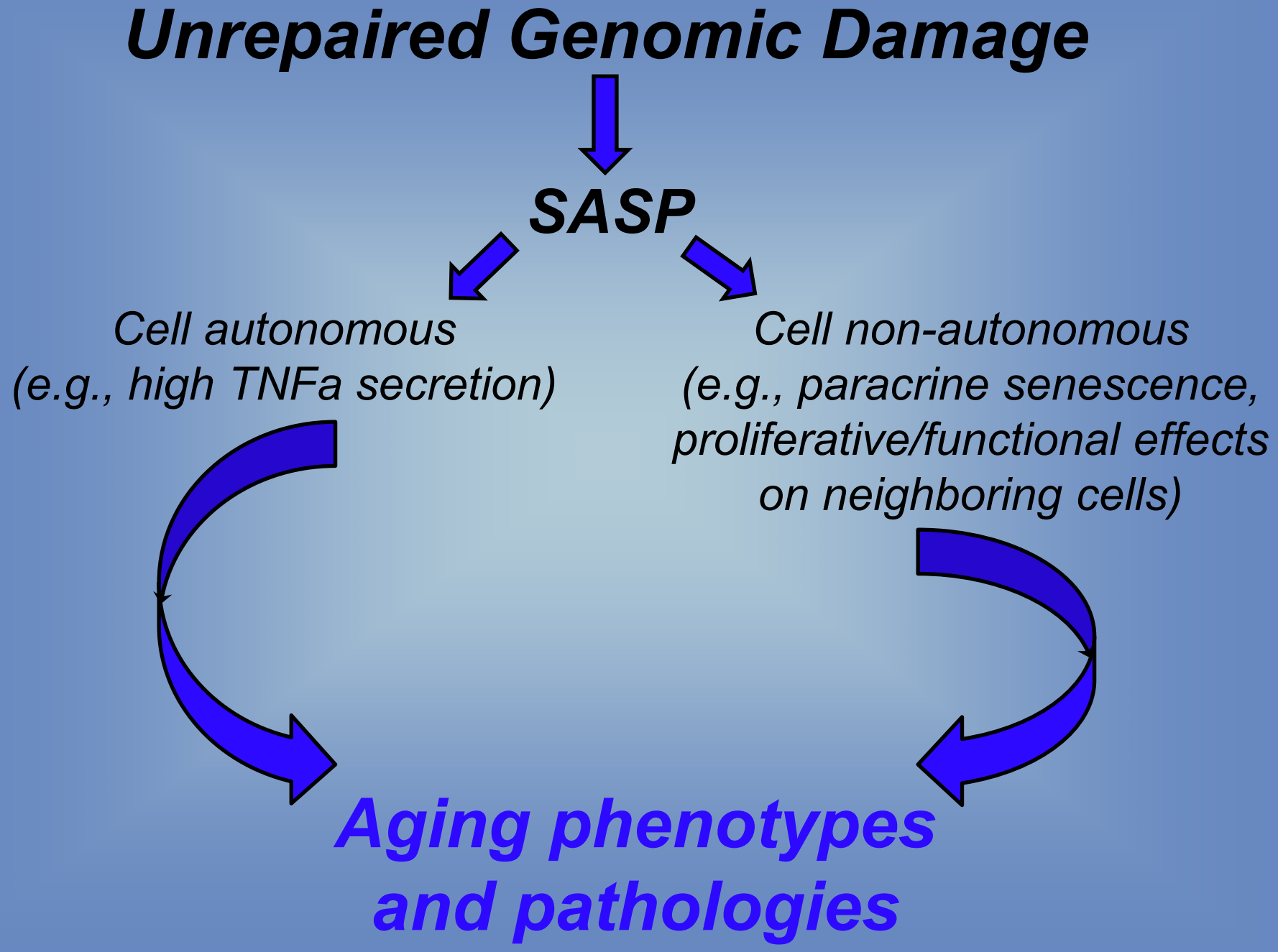
Basisty et al, PLoS Biol, 2020

The SASP is dynamic





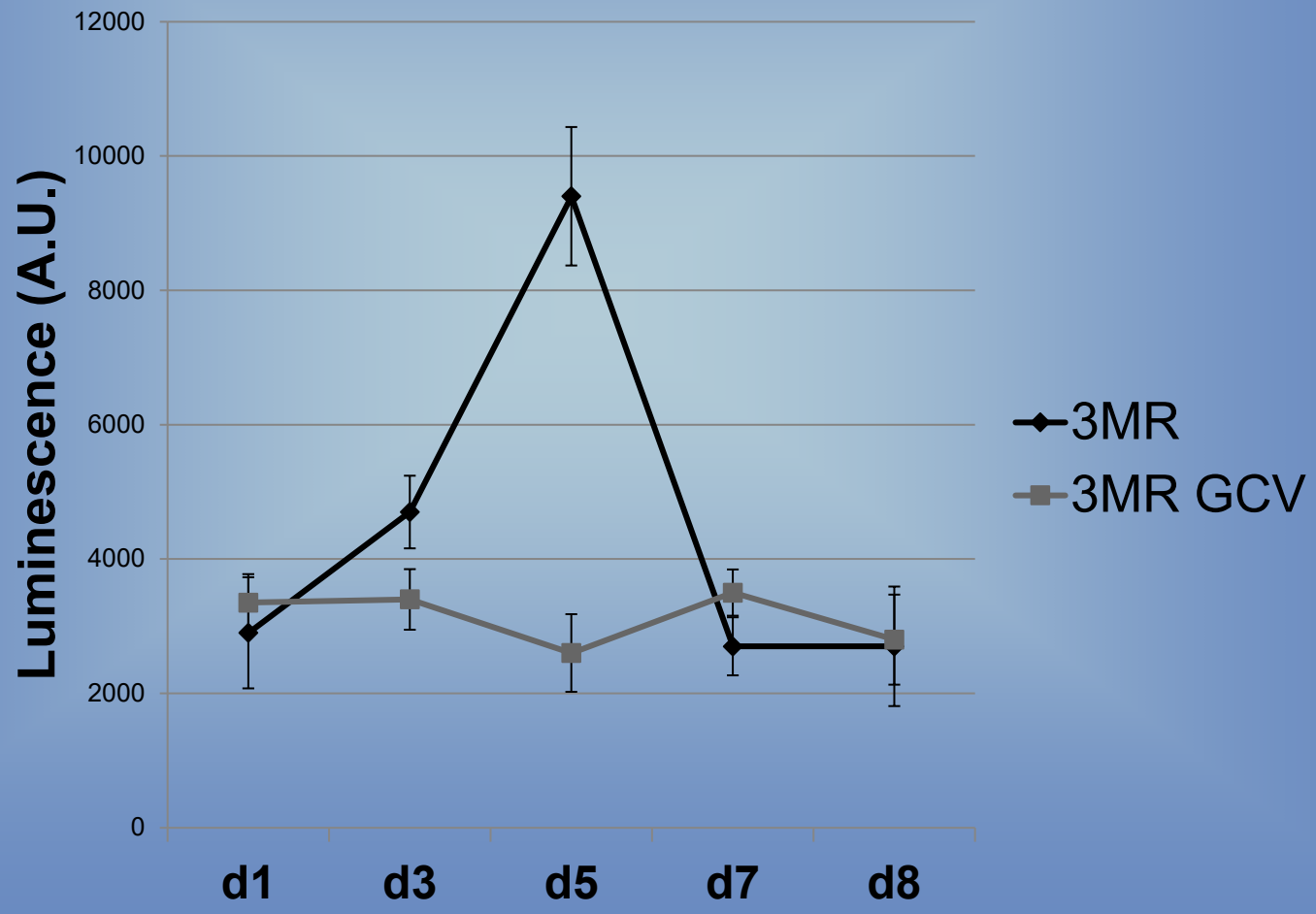
Wiley, Davis, Ramanathan, LeSaux; JCI Insight, 2019


Unrepaired Genomic Damage
SASP
Cell autonomous
(e.g., high TNFa secretion)
Cell non-autonomous
(e.g., paracrine senescence,
proliferative/functional effects
on neighboring cells)
Aging phenotypes
and pathologies

Cellular senescence and wound healing

(why did the SASP evolve?)

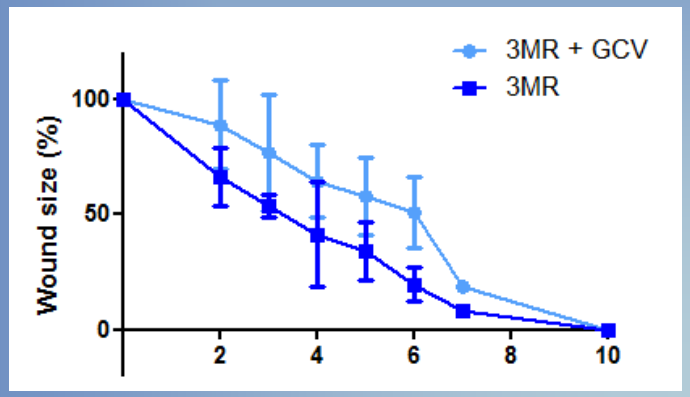
Senescent cells are present transiently during wound healing

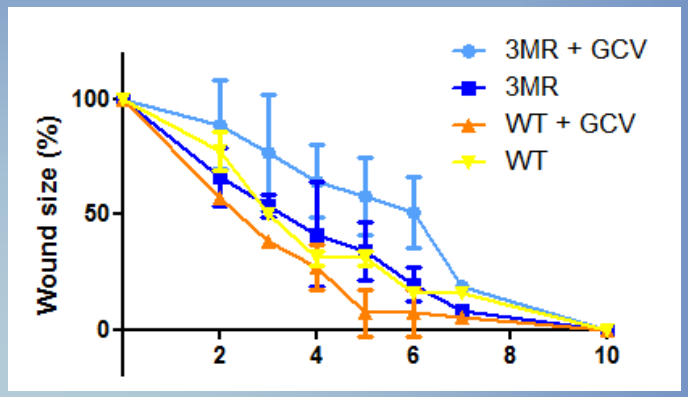


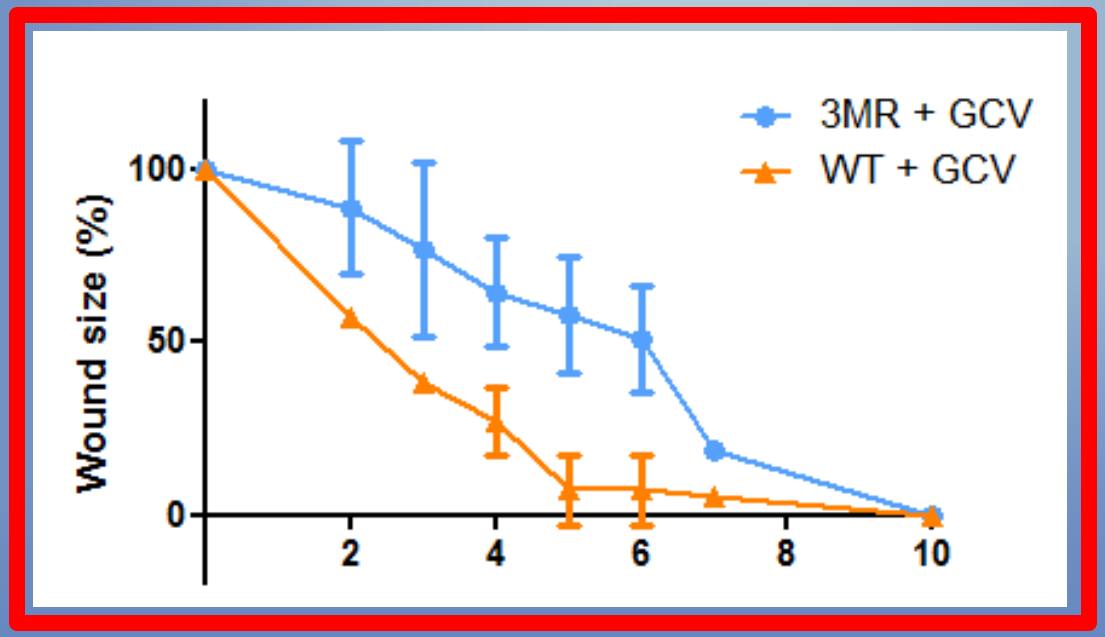
female mice

WOUND HEALING IS RETARDED BY ELIMINATING SENESCENT CELLS

GCV 0-5 days after biopsy







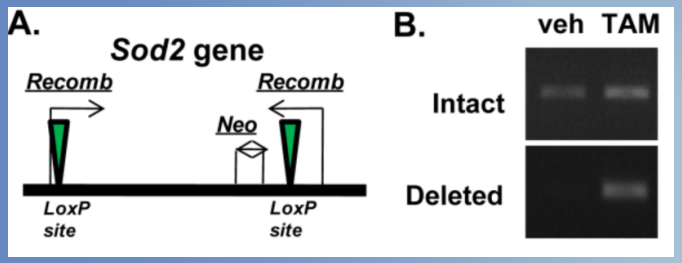
SnCs promote wound healing largely by secreting PDGF-AA

but ….

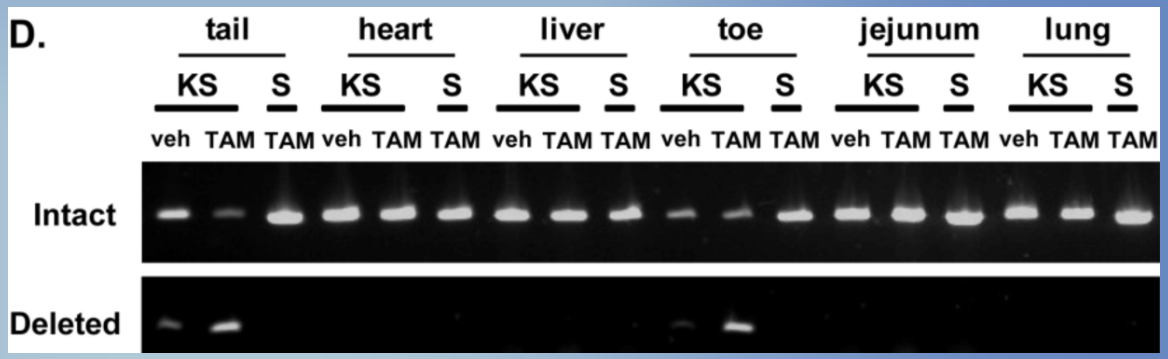
senescent cells accumulate with age
and
wound healing slows with age

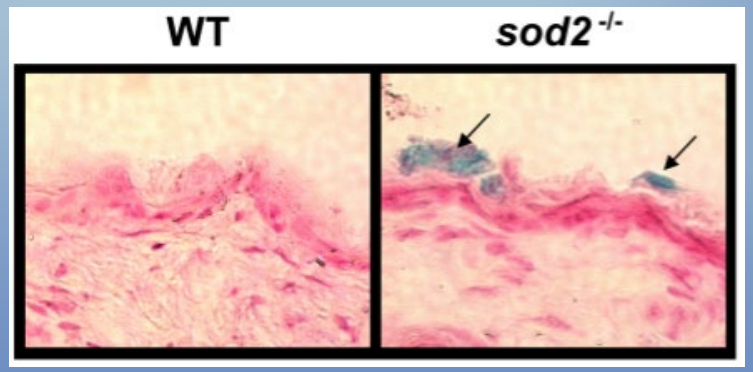
WHY?

Epidermal-specific deletion of SOD2 causes persistent senescence in the skin



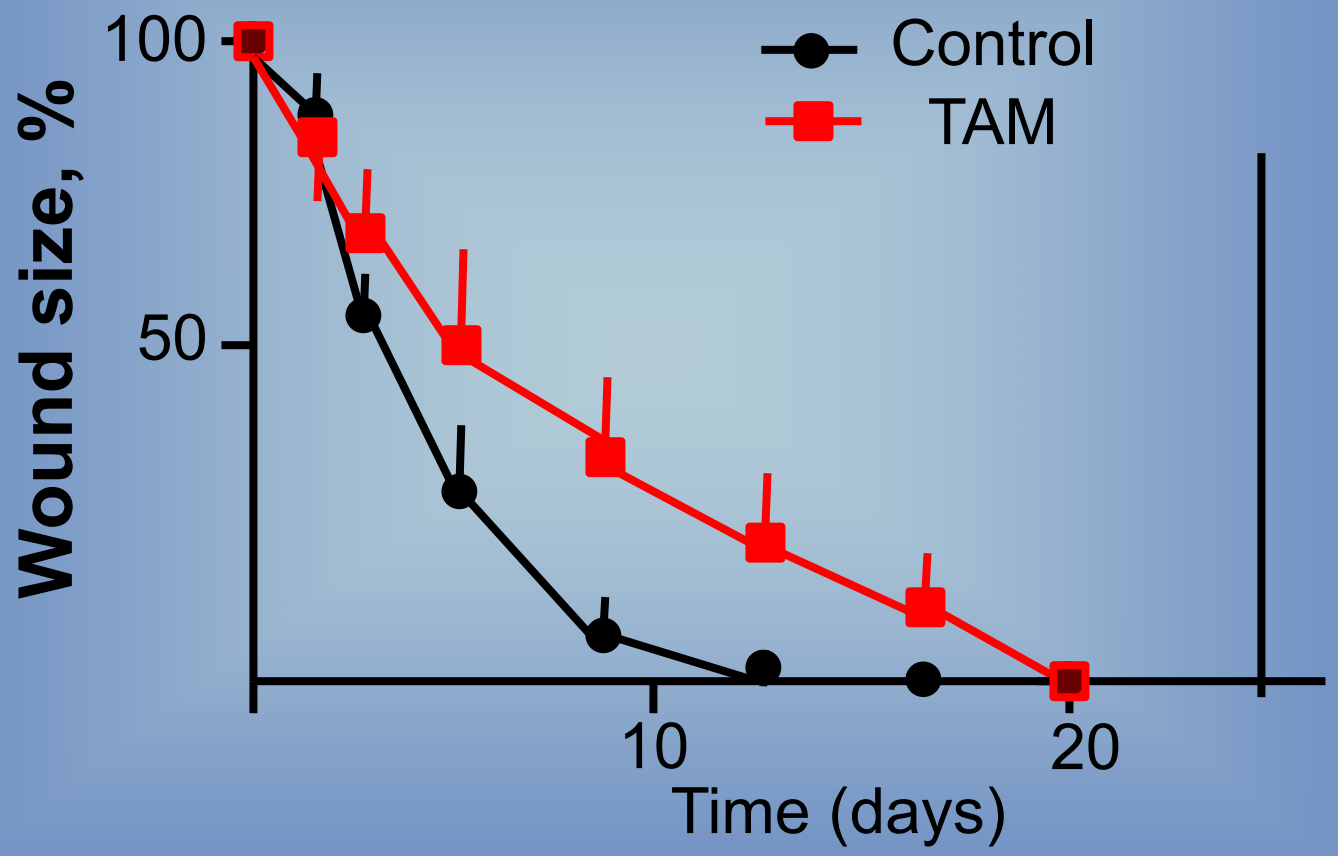
x KTR-Cre(ERT)





Velarde et al, Aging, 2012; Velarde et al, PNAS, 2015

…. and retards wound healing



Velarde et al, PNAS, 2015; Aging 2012

male mice

Transient presence of senescent cells are required for optimal tissue repair

inflammation

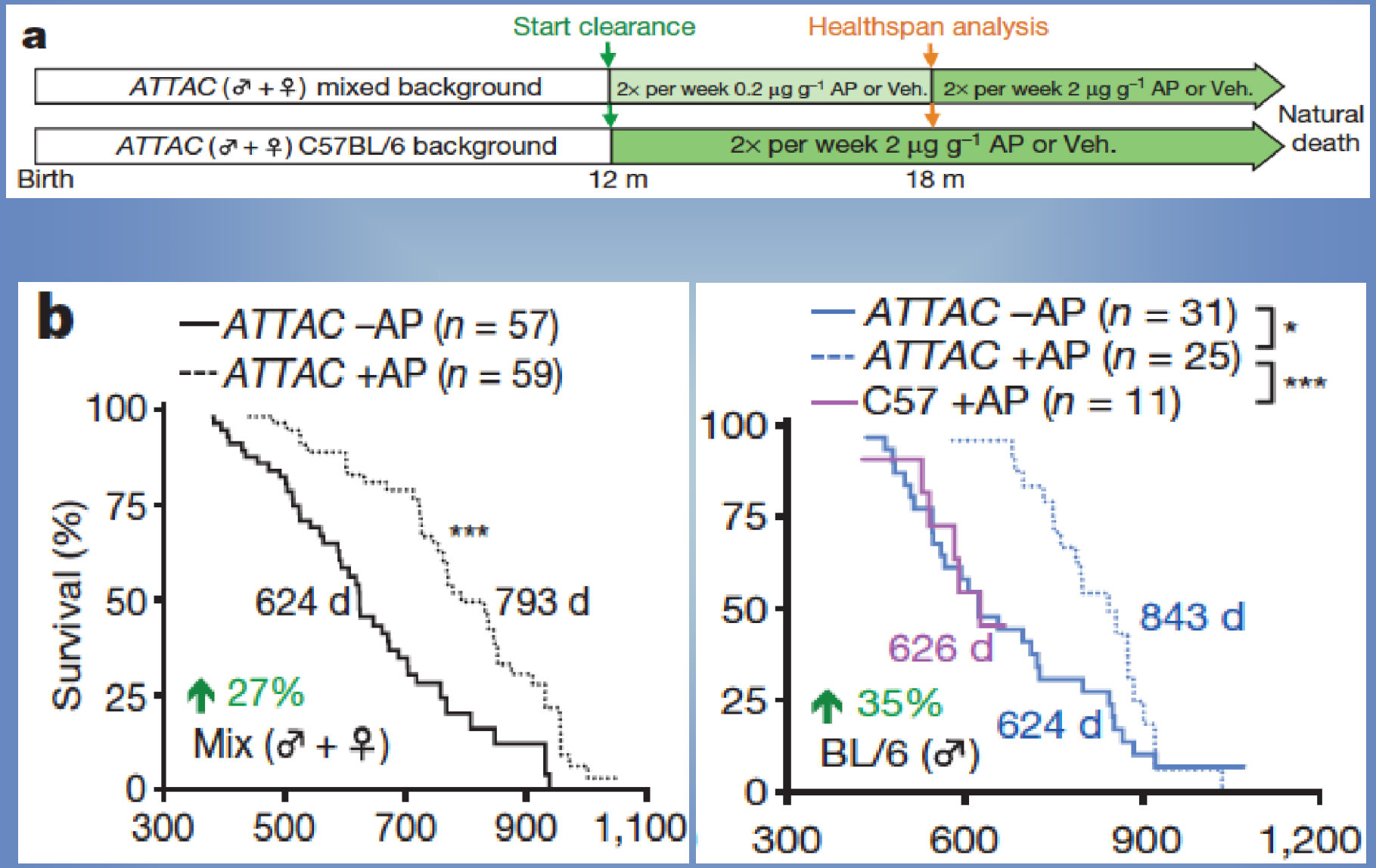
proliferation, granulation

matrix formation/remodeling

Persistent senescent cells induced by stressors (chemo, ox stress, aging) retard tissue repair

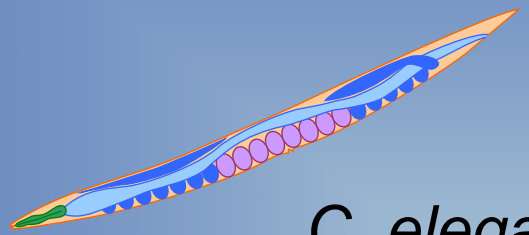
'Senolytic' and 'senomorphic' drugs are on the horizon, and hold promise for extending health span!

Do they hold promise for extending life span?



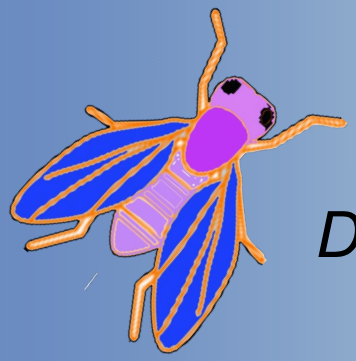
Baker et al, Nature, 2015

Life span extension …..



C. elegans

10-fold!



Drosophila

<2-fold

Mouse

5-20%

THANKS!

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