

MODULE 3

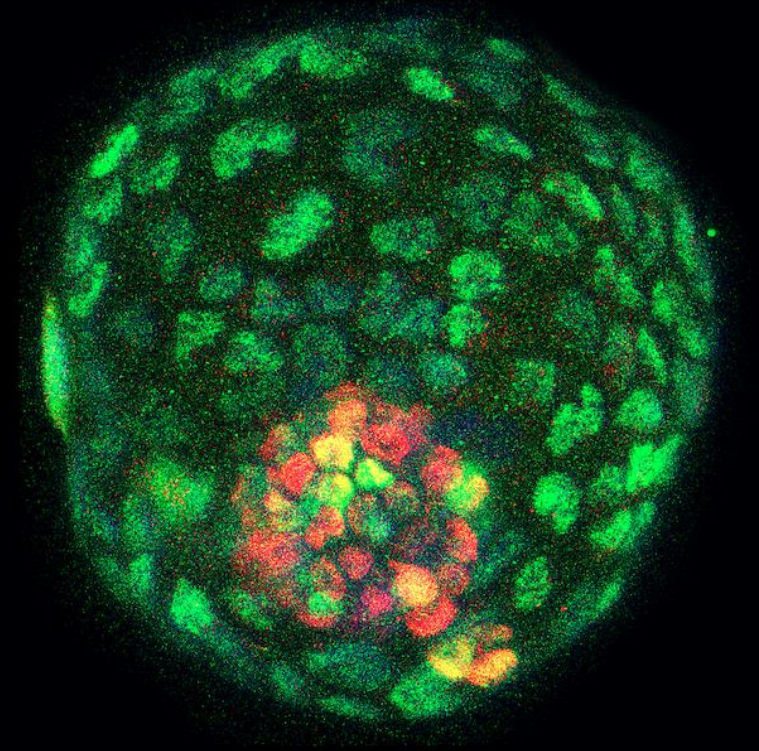
INTRODUCTION TO HUMAN CELL MODELS OF AGING CORE

- SD-NSC HUMAN COHORT
- CELLULAR MODELS OF AGING

Co-Leaders:

Anthony Molina (UCSD)

Rusty Gage (Salk)

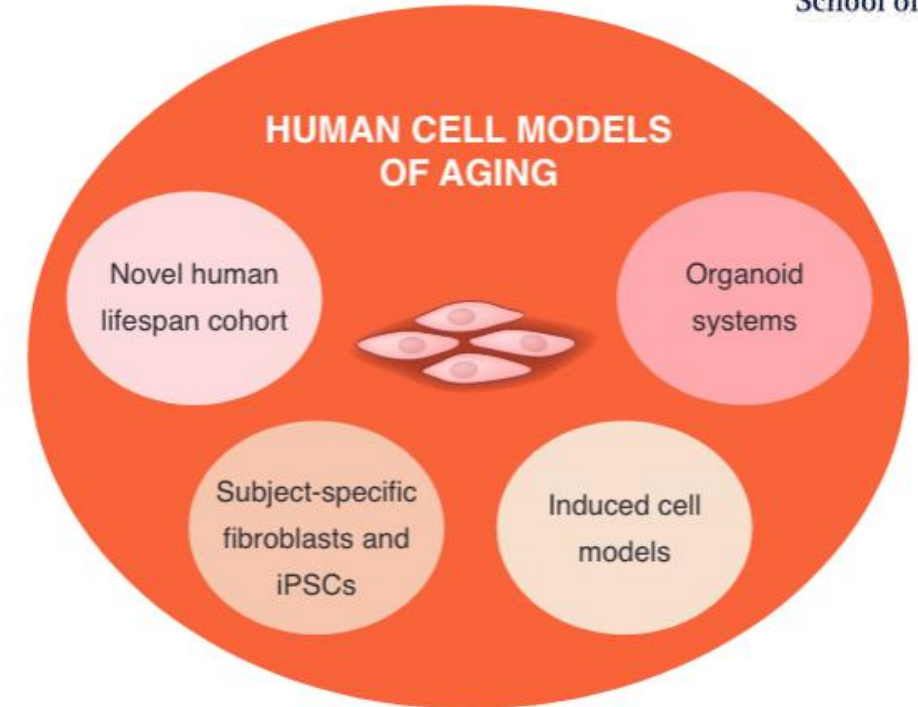


INTRODUCTION TO HUMAN CELL MODELS OF AGING CORE

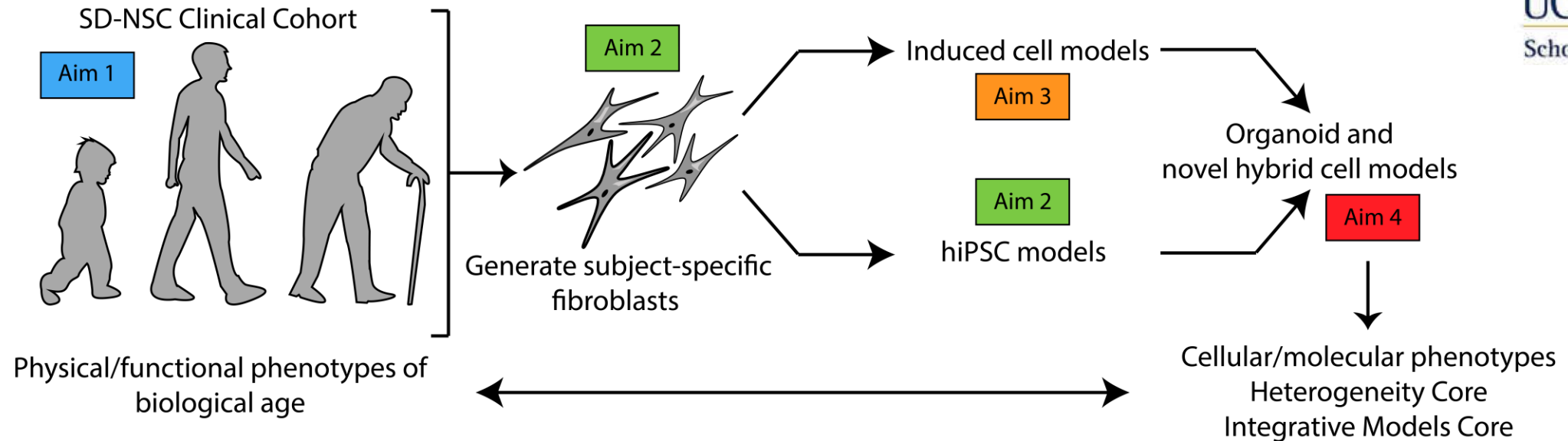


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- INTRODUCTION TO HUMAN CELL MODELS OF AGING CORE
- OVERVIEW OF **NOVEL HUMAN LIFESPAN COHORT**
- OVERVIEW OF **FACILITY RESOURCES**
- OVERVIEW OF **CELL MODELS**

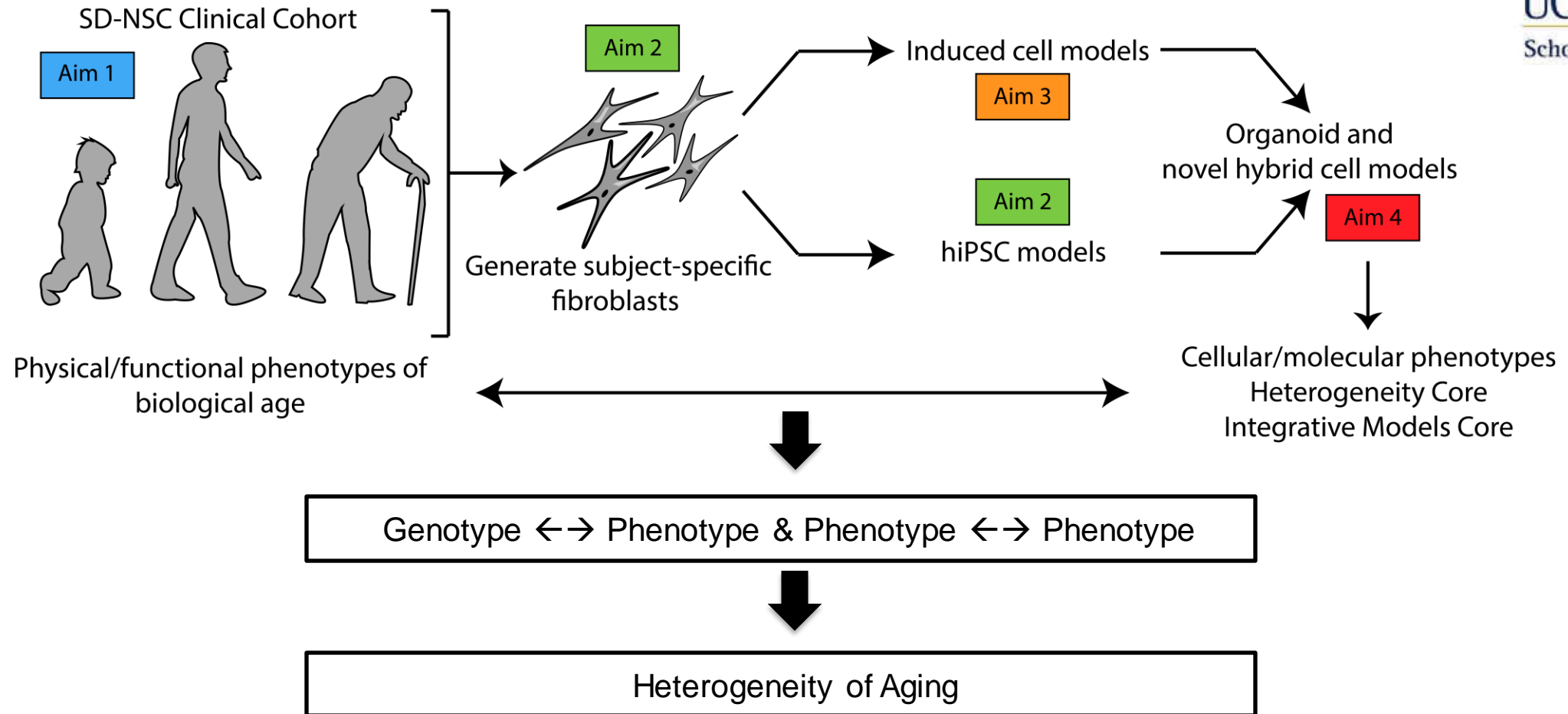


WELL PHENOTYPED COHORT POWERS NOVEL CELL MODELS



- **SD-NSC Cohort will represent the adult human lifespan** and be extensively phenotyped for physical and functional metrics of biological aging
- Subject-specific fibroblast used to generate ***iCell models (capture age related phenotypes)*** and hiPSCs
- hiPSCs used to generate ***niche specific multi-cellular organoid models***
- ***iCell and hiPSC models combined*** into novel hybrid models of human aging

WELL PHENOTYPED COHORT POWERS NOVEL CELL MODELS



What is “representative” or “normal” when it comes to aging?

Major Considerations:

- Healthy vs Normal
- Majority of patients over 65 present with multiple comorbidities

Inclusion	Exclusion
<ul style="list-style-type: none">-Over 20yrs of age-Able to consent and participate in the study using English-BMI ≥ 18.5 and ≤ 30 kg/m².-Weight stable for the prior 6 weeks-Normal cognitive function-Willing and able to attend two in-person study visits that will include vigorous exercise testing, blood draw, and skin biopsy.-Willing to wear a wireless accelerometer (Actigraph GT3X) for 14 days	<ul style="list-style-type: none">-Are pregnant-Diabetes (fasting glucose >126 mg/dl)-Uncontrolled hypertension (BP $> 140/90$ mmHg)-Heart or cardiovascular condition, including coronary artery disease, congestive heart failure, diagnosed abnormality of heart rhythm, atrial fibrillation, and/or a history of myocardial infarction-Cancer or history of cancer-Dementia or other conditions that may affect cognitive ability-Sensory or physical impairment that would prevent participation-Parkinson's disease, multiple sclerosis, or other neurological condition, including a previous stroke, which may be causing impaired muscle function or mobility-Medications and supplements that may interfere with measurements or biological outcomes including, but not limited to: metformin, CoQ, glucocorticoids, and medications that may alter cardiac and hemodynamic responses to exercise-Respiratory disease-Answers “yes” to one or more questions in the American College of Sports Medicine’s Physical Activity Readiness Questionnaire (PAR-Q) and/or report two or more risk factors for exercise testing

Defining Biological Age

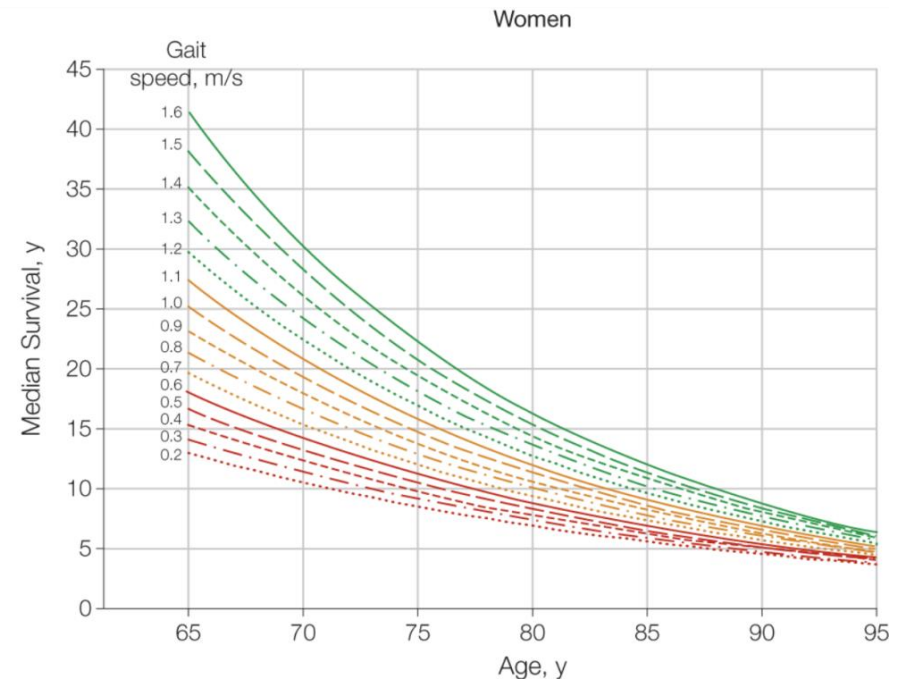
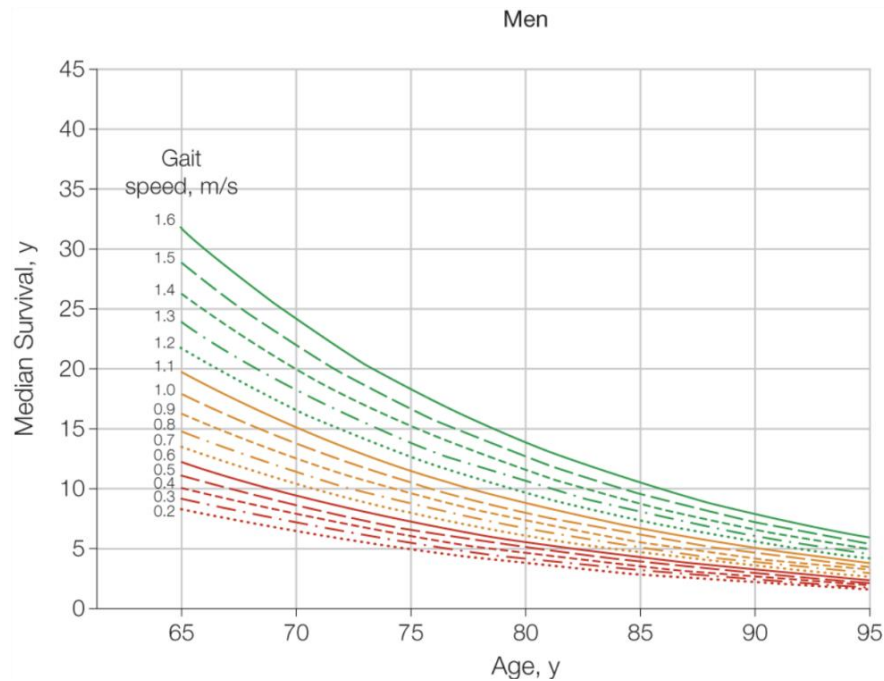
-Physical function is among the best predictors of morbidity and all cause mortality



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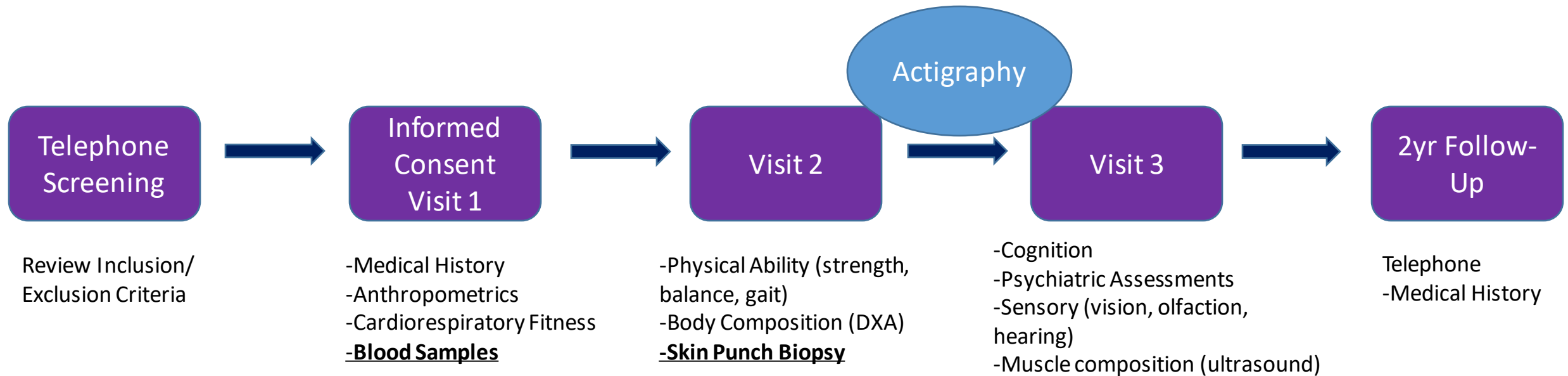
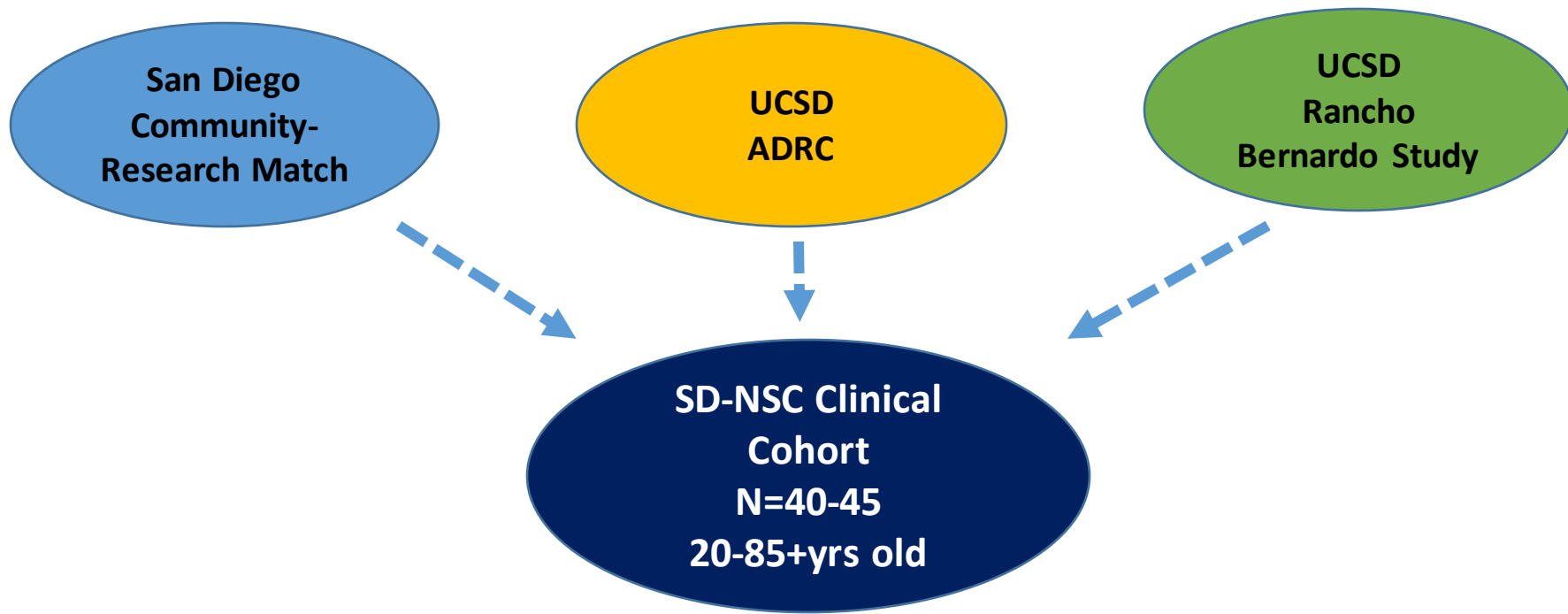
Pooled data from 9 cohort studies
34,485 total participants, 17 528 deaths

-Remaining years of life plotted by age group



10 yr survival Hazard ratio for every 0.1 m/s was 0.88

Studenski JAMA, 2011

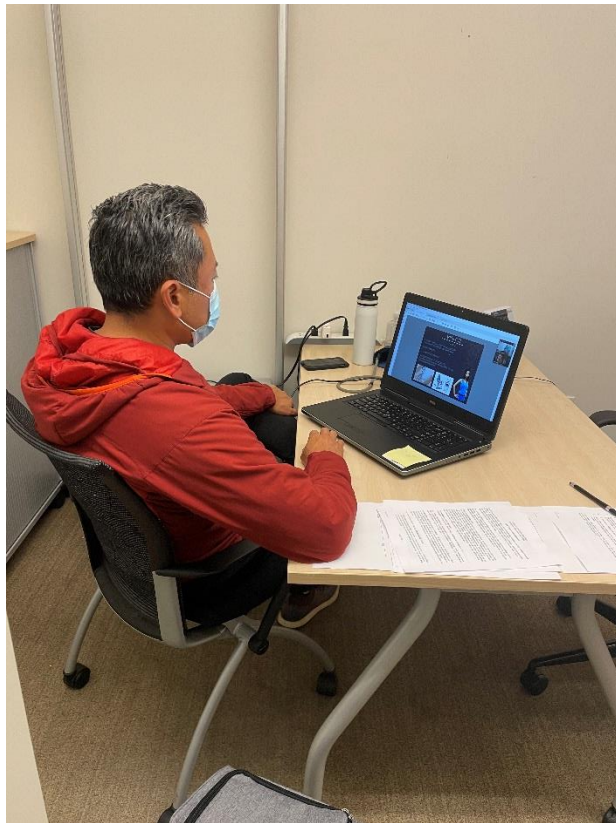


Visit 1

salk
Where cures begin.

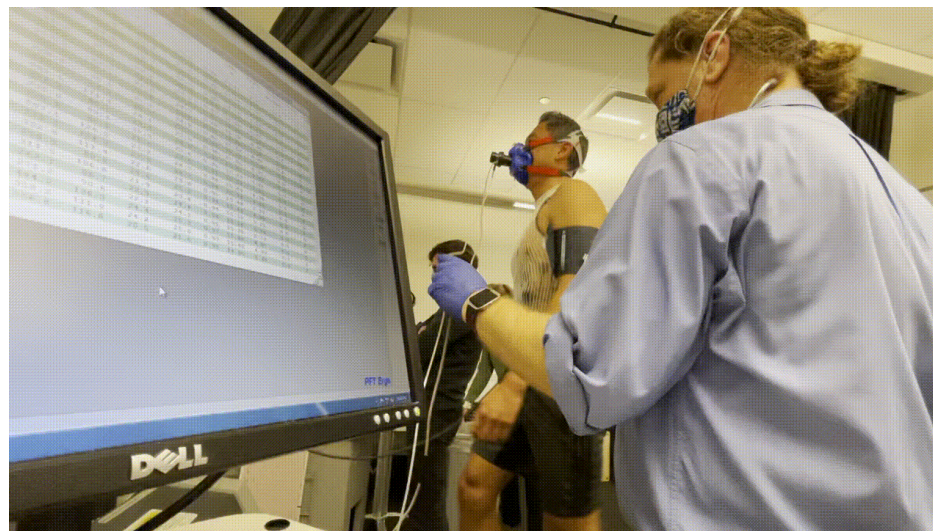
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Informed Consent During the Covid-19 Pandemic



Blood Sample Collection





-VO2 max is generally regarded as the best indicator of cardiorespiratory fitness.

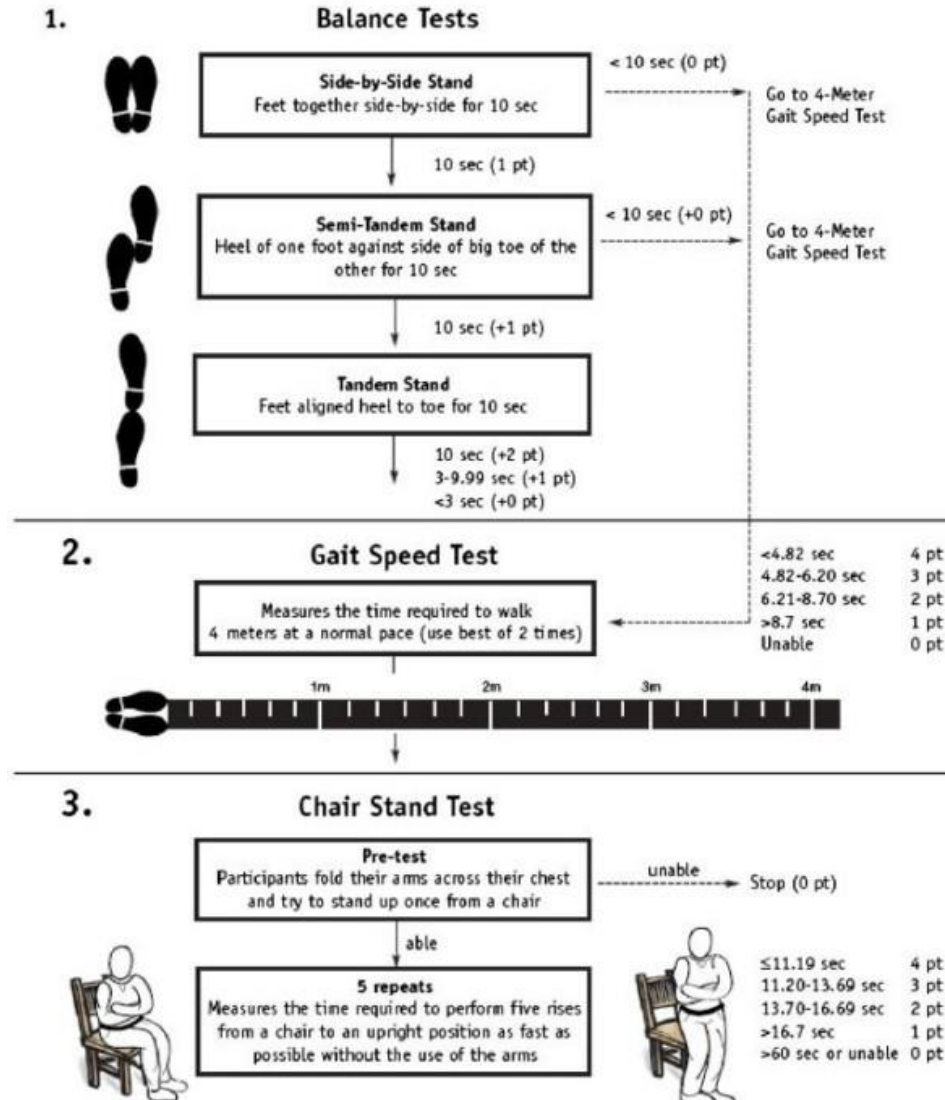
-Safety

- Persons with contraindications will be excluded from the study
- Monitoring (Physician, Exercise Physiologists)
 - EKG, Blood Pressure

-Age/Ability appropriate

- Exercise Modalities

Limitations
-Ceiling Effects



Visit 2

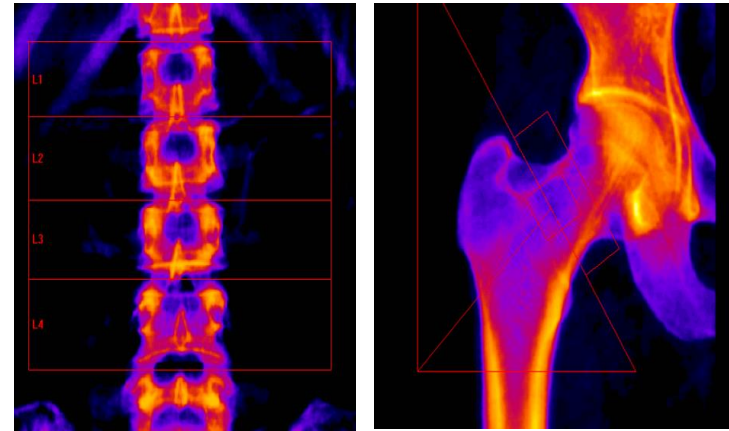
Dual-energy X-ray absorptiometry (DXA)

salk
Where cures begin.

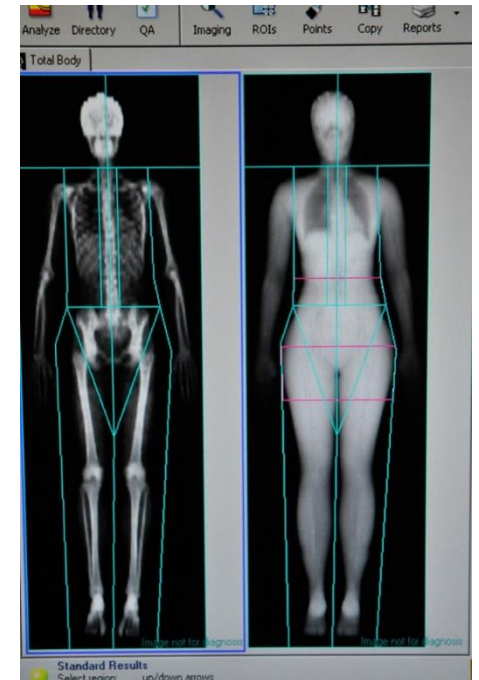
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Bone Mineral Density



Body Composition



Visit 2

Physical Performance

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Where cures begin.

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Gait Speed

- 6mwd (fast)
- 2.5 mwd (usual)



Leg Strength (Biodex)



Grip Strength



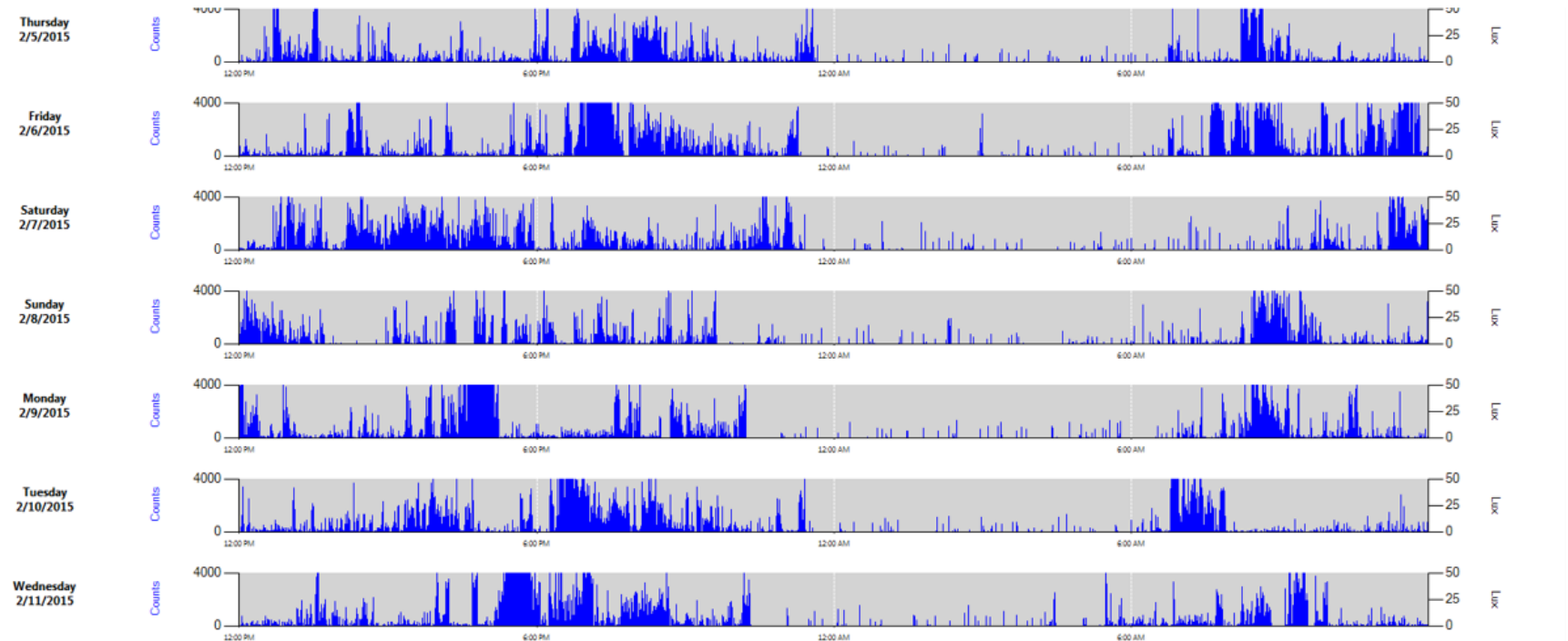
Visit 2-3

Actigraphy

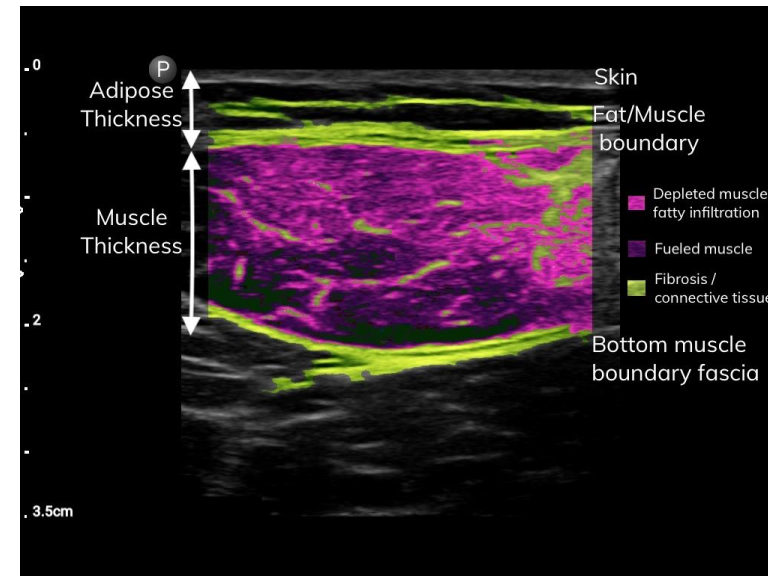
monitoring rest/activity cycles
2 weeks

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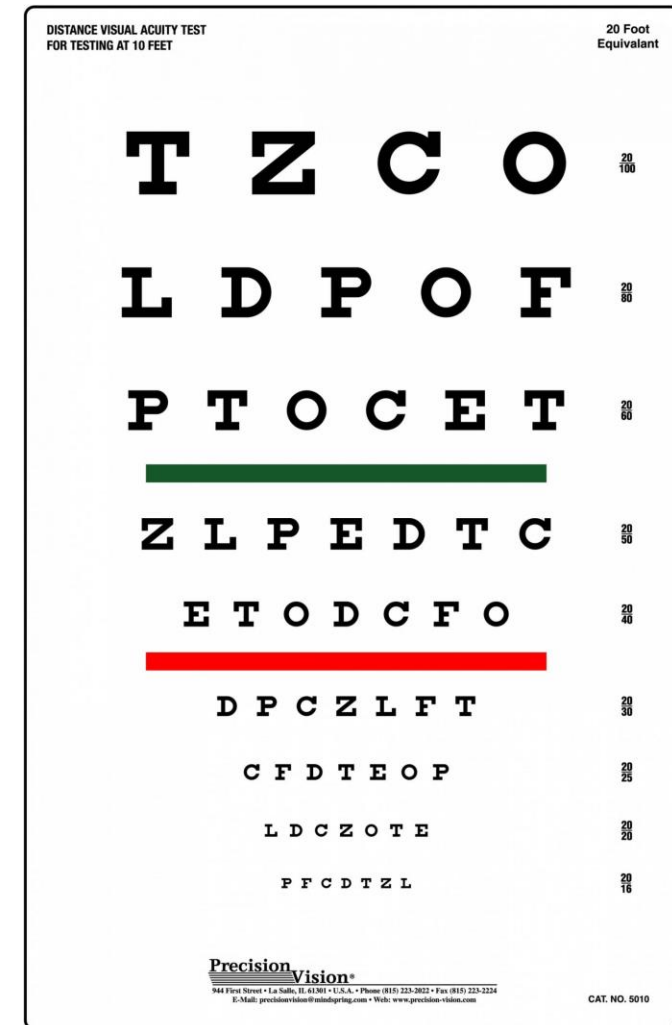


Size and Adiposity



Visit 3

Sensory Abilities





Biological Samples

Limitations of SD-NSC Cohort



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SD-NSC Biorepository
Rolling Enrollment (2021-2023)

Blood

- Platelets

- WBCs

- Plasma

Cells

- Primary dermal fibroblast

- reprogrammed iPSCs (stem cells)

- Small Size (N=40)

- Inclusion/Exclusion Criteria

- Diversity

 - Sex

 - Race/Ethnicity

- Suitability of measures for broad age range

***Opportunities to expand**

- # participants

- Representation

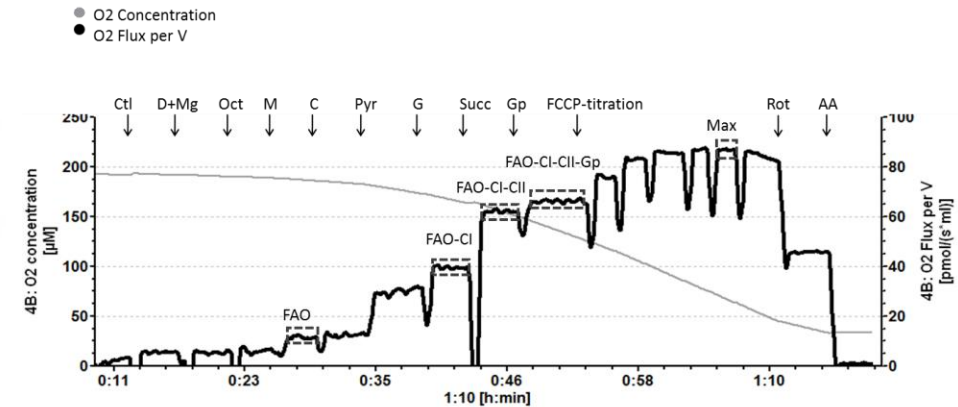
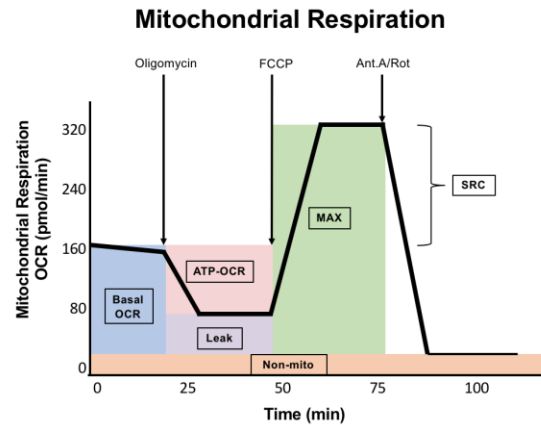
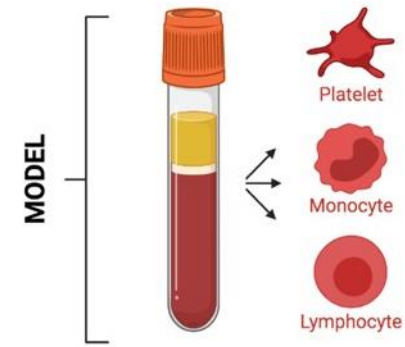
- Measurements

- Biospecimens

- Longitudinal Follow Up**

**Based on future grant funding*

Mitochondrial Bioenergetics – Blood Cells



Agilent Seahorse XF Analyzer

- High Throughput
- Intact cells
- 4 injection ports

Oroboros Oxygraph-2K

- High resolution
- Intact and permeabilized cells/tissues
- Unlimited injections

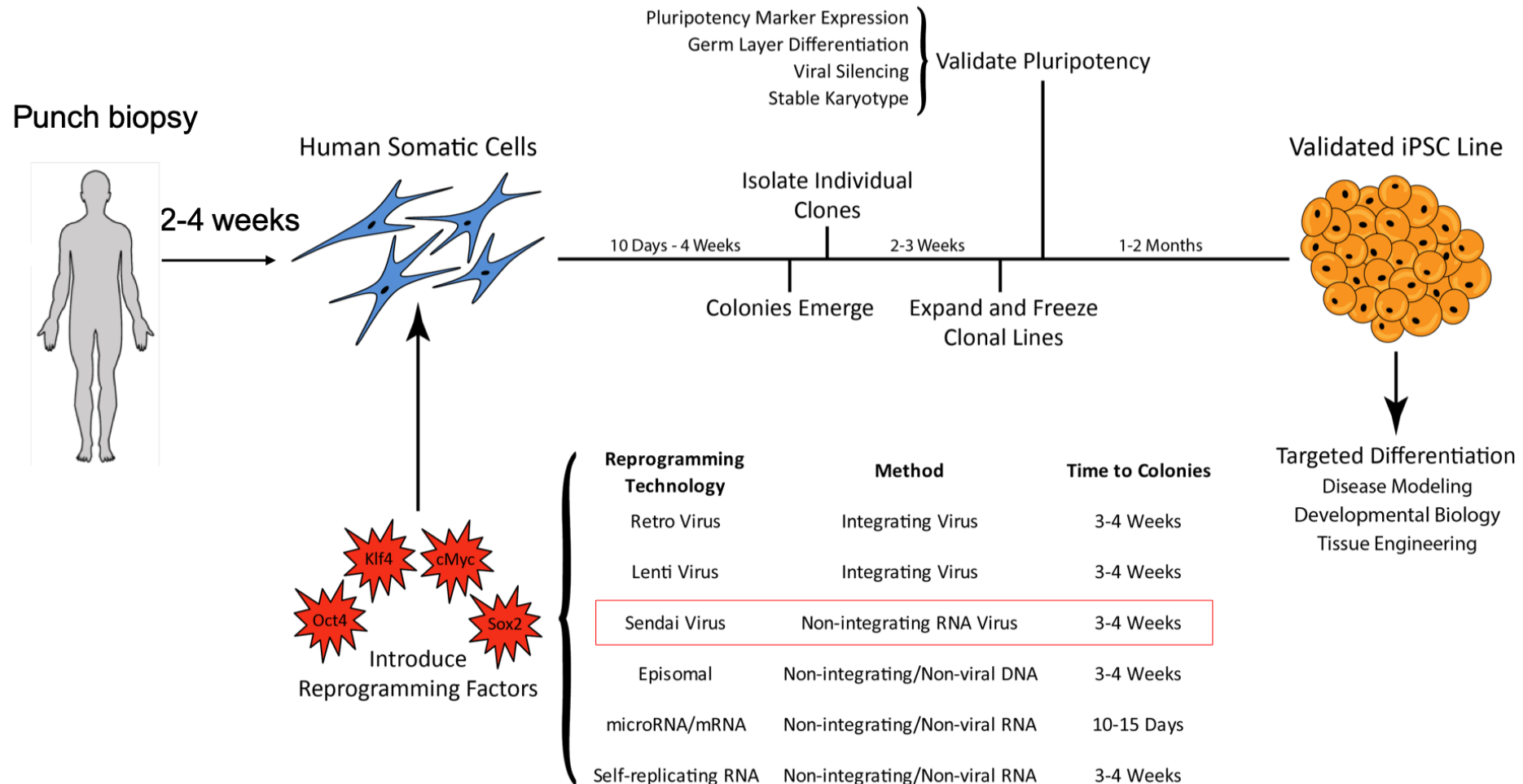
Skin Punch Biopsy



**SD-NSC Cell
Models Core @Salk**

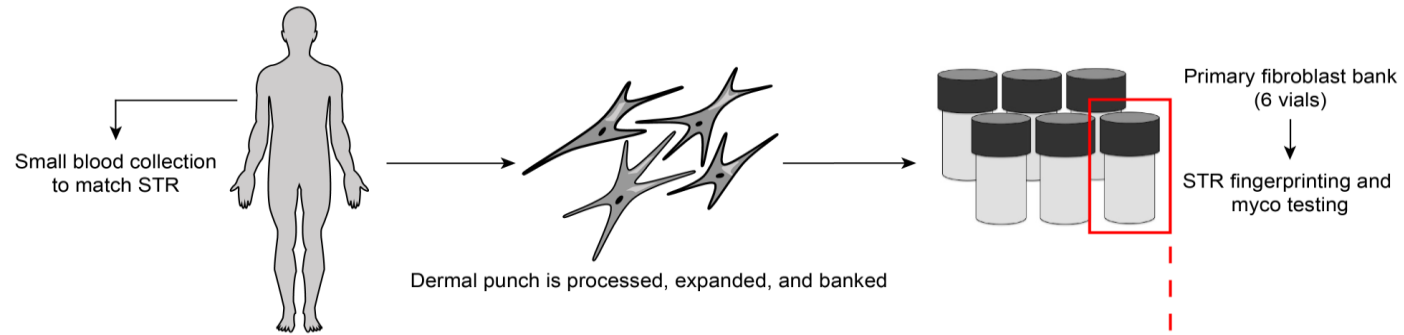
WELL VERSED IN BEST PRACTICES FOR HUMAN CELL MODELS

Derive → Bank → Characterize → Model

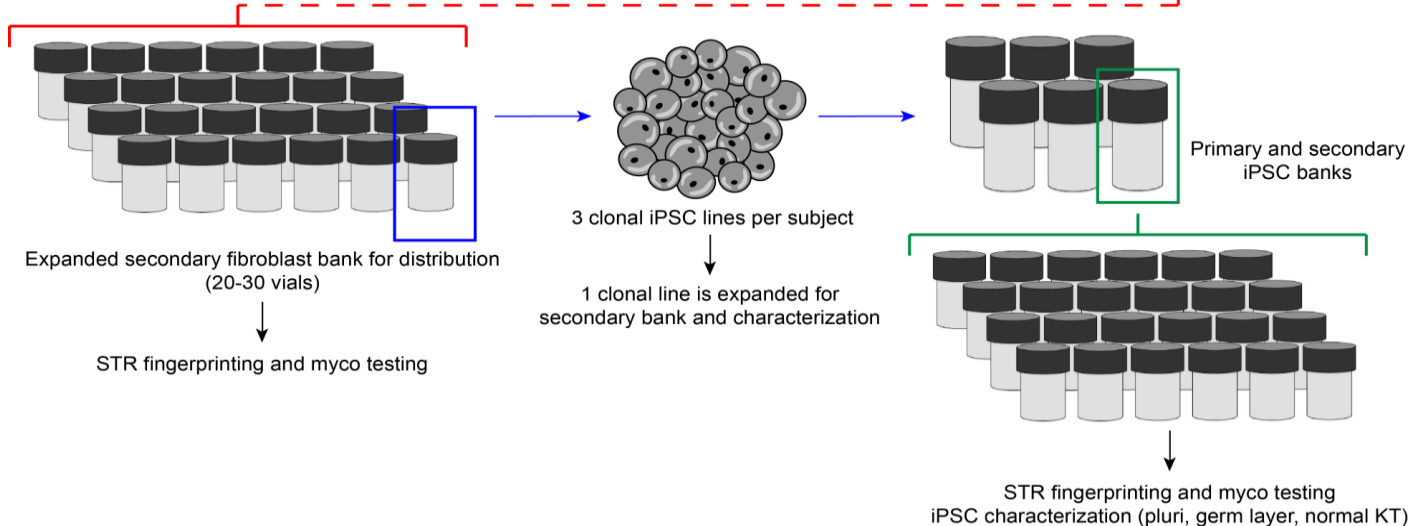


Tiered Banking Ensures Prolonged Access to Cell Resources

Tier 1 - Process punch biopsy and establish primary cell bank



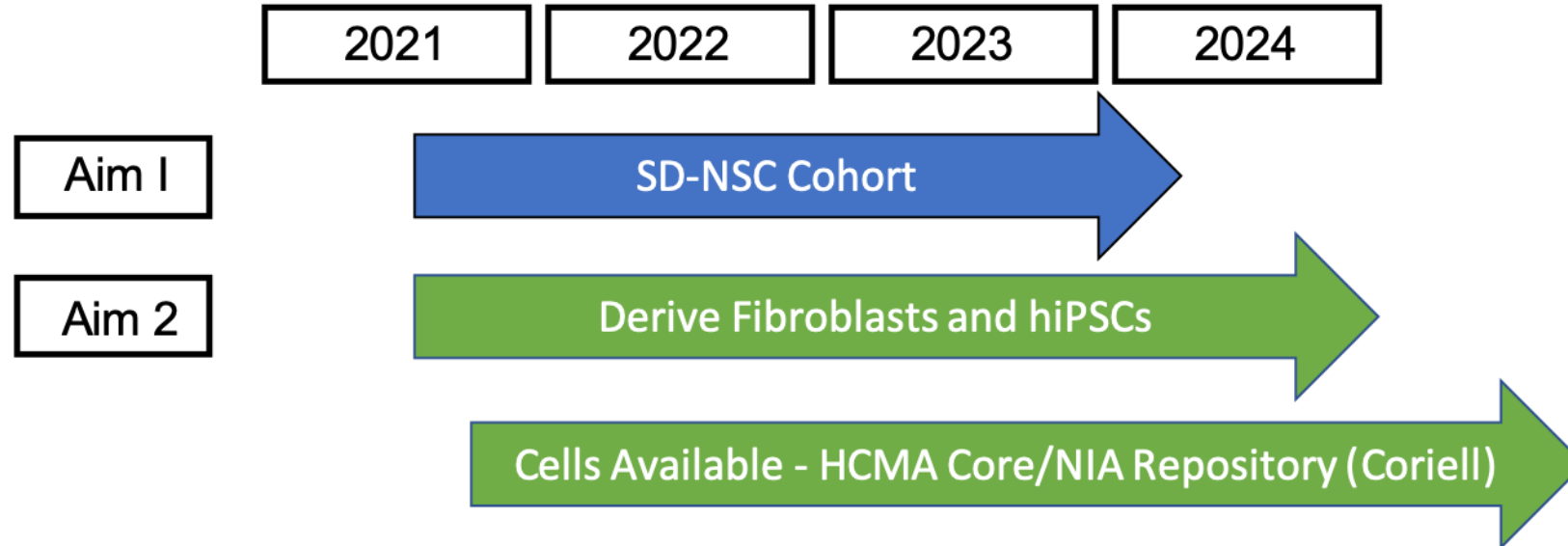
Tier 2 - Establish secondary fibroblast bank and reprogram to



ESTIMATED TIMELINE FOR CELL RESOURCES



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Fibroblasts and hiPSCs available in late 2021

Early sharing through HCMA Core (SD-NSC Website)

Plan to deposit with national repository for broader distribution (NIA Repository at Coriell)

Protocols for basic fibroblast/hiPSC maintenance available mid 2021

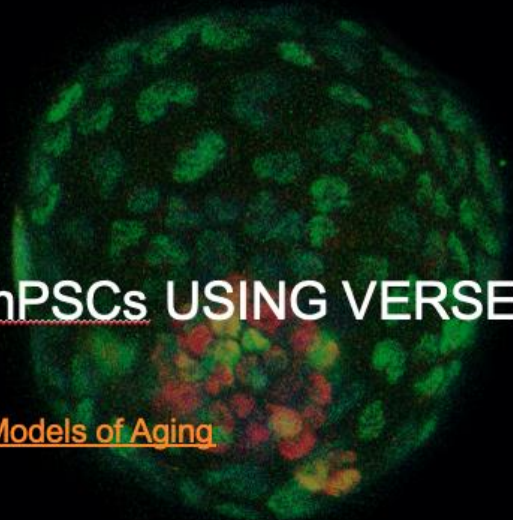
Formal written and image-enhance protocols (SD-NSC Website)

SAMPLE OF IMAGE-ENHANCED PROTOCOLS




Coming soon to the SD-NSC Website

**SAN DIEGO NATHAN SHOCK
CENTER**

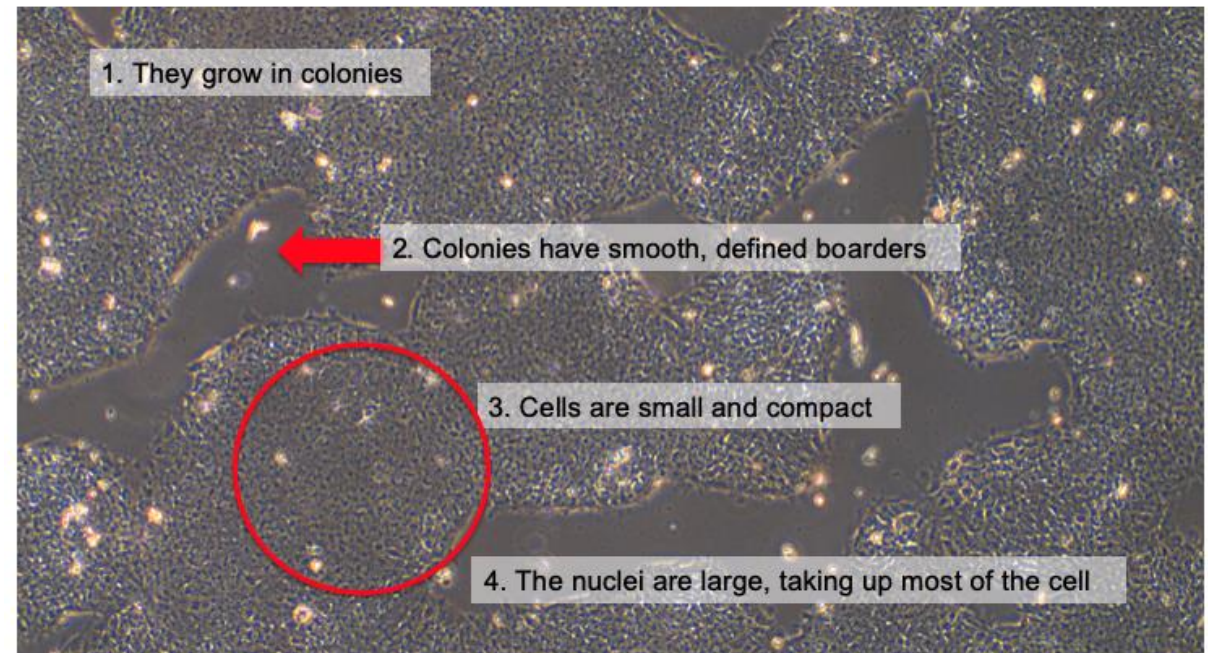


**PASSAGING hPSCs USING VERSENE
(EDTA)**
SD-NSC Human Cell Models of Aging
Published: March 2021
Research reported in this publication was supported by the National Institute On Aging of the National Institutes of Health under Award Number P30AG068635

**WWW.SALK.EDU**

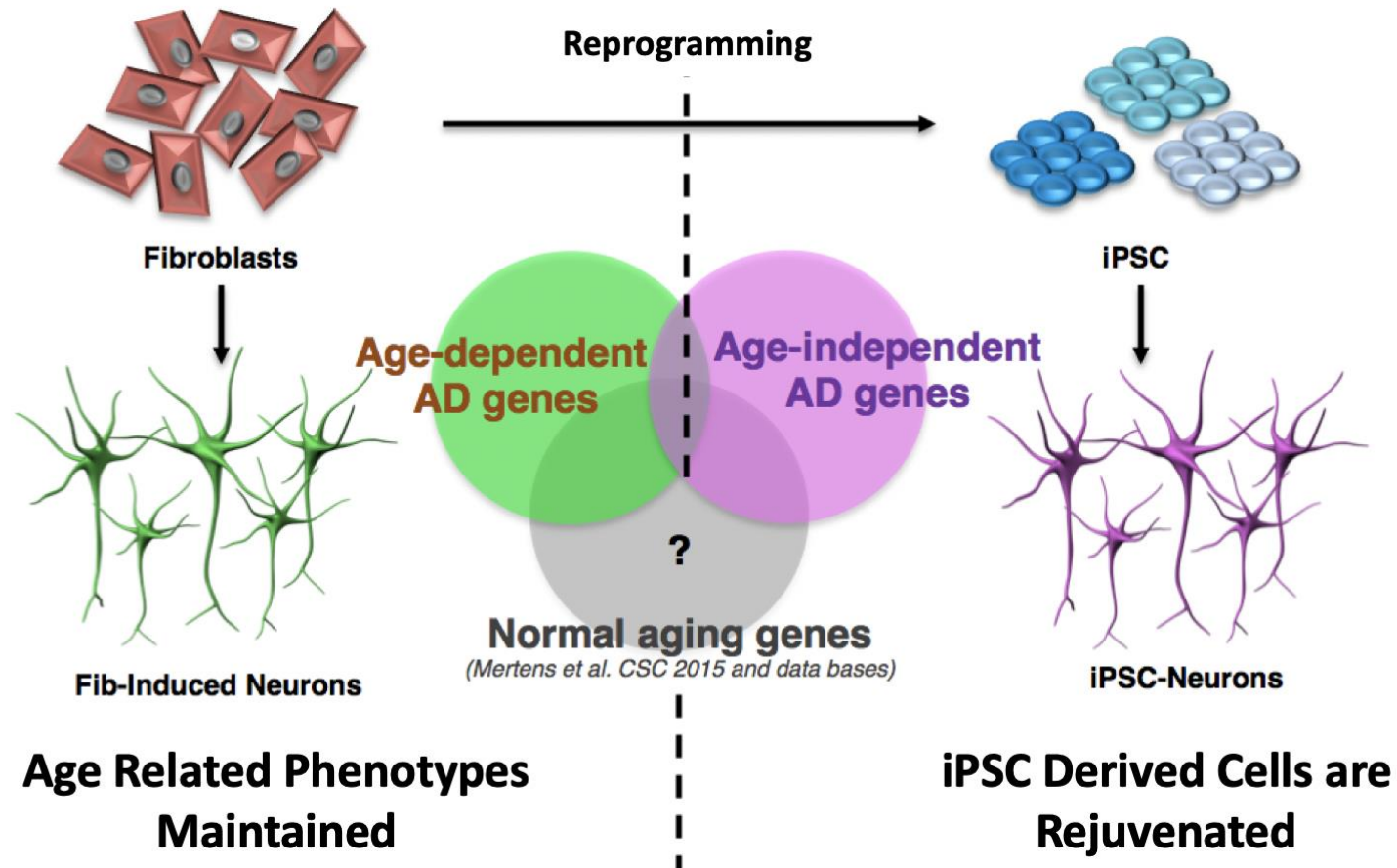
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Healthy hPSCs have 4 key characteristics

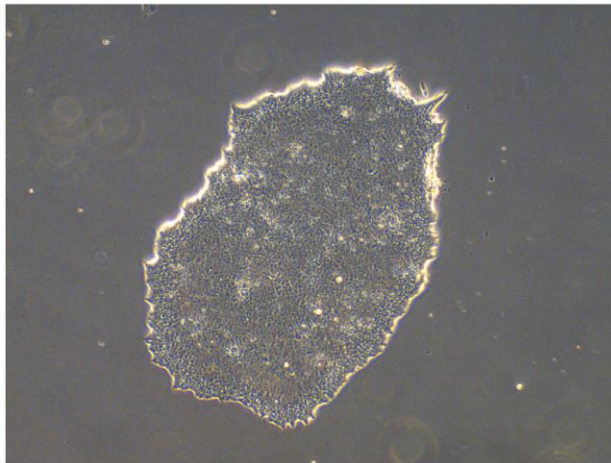


SALK INSTITUTE FOR BIOLOGICAL STUDIES

iCell Models Maintain Age Related Phenotypes



hiPSCs Recreate Niche Specific Dynamics Through Organoid Models

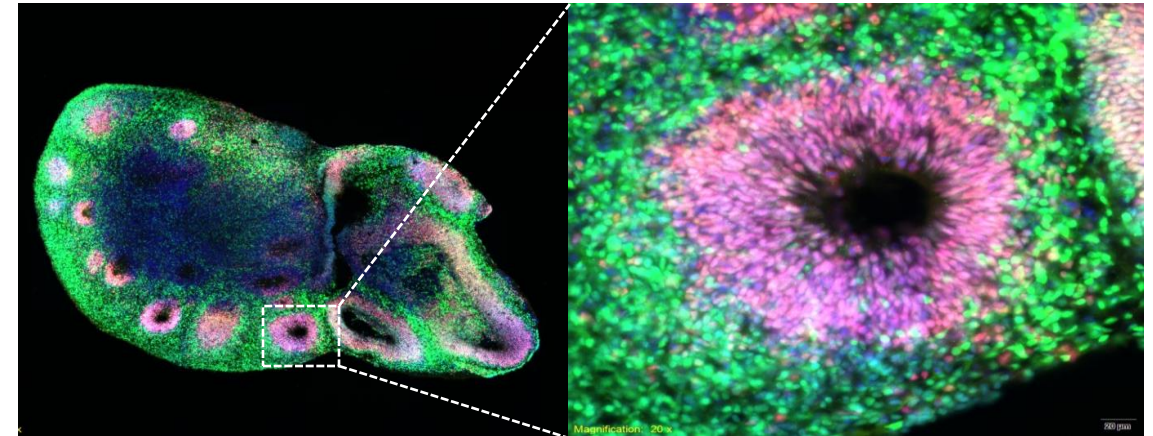


2D hiPSCs

3D, Matrix, Morphogens

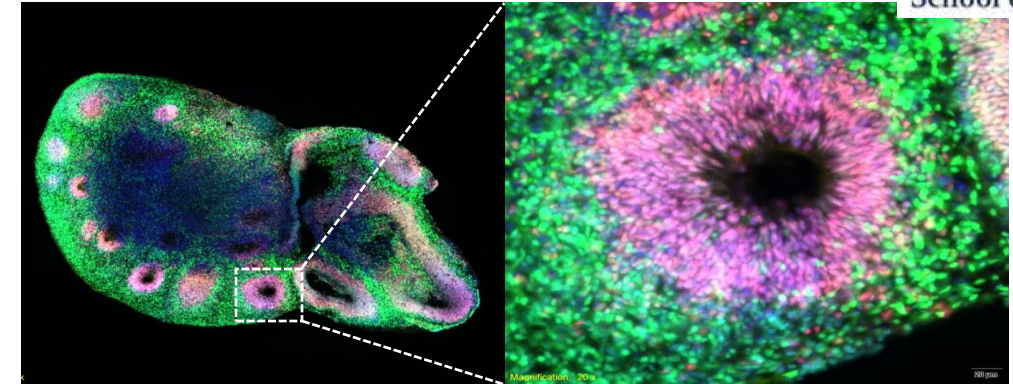
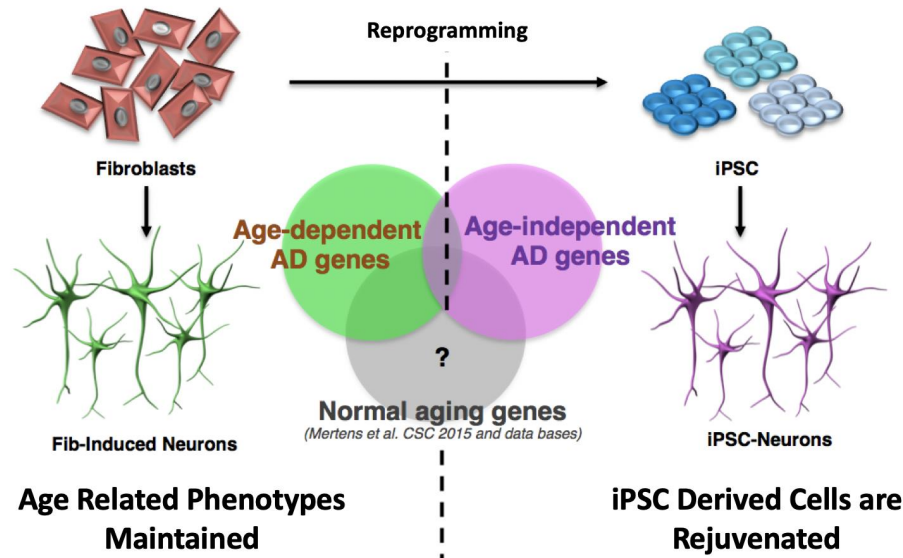


Time



3D Organoids w/Multi-Cellular
Structural Complexity

HYBRID MODELS COMBINE THE BEST OF BOTH WORLDS



Niche Specific Multi-Cellular Dynamics

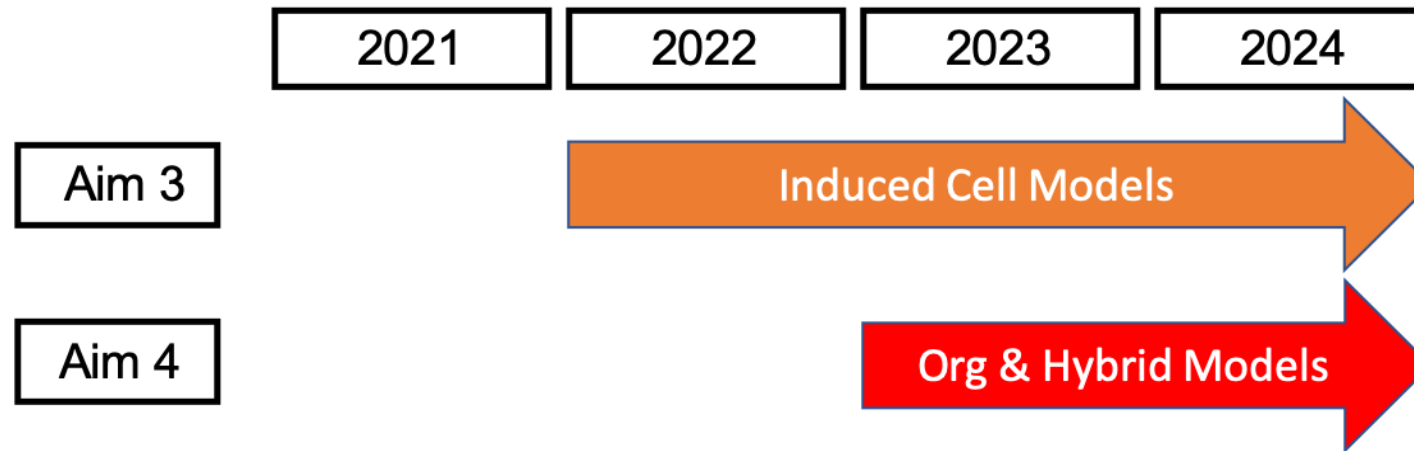
Age Related Phenotypes

Hybrid Models of Human Aging
Aged Niche in a Dish

ESTIMATED TIMELINE FOR CELL MODELS



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Induced cell models available in 2022

Neurons (iN) and Vascular Endothelial Cells (iVECs)

Detailed Protocols (SD-NSC Website)

Updates made available (SD-NSC Website)

Organoid and Hybrid Cell Models in 2023-2024

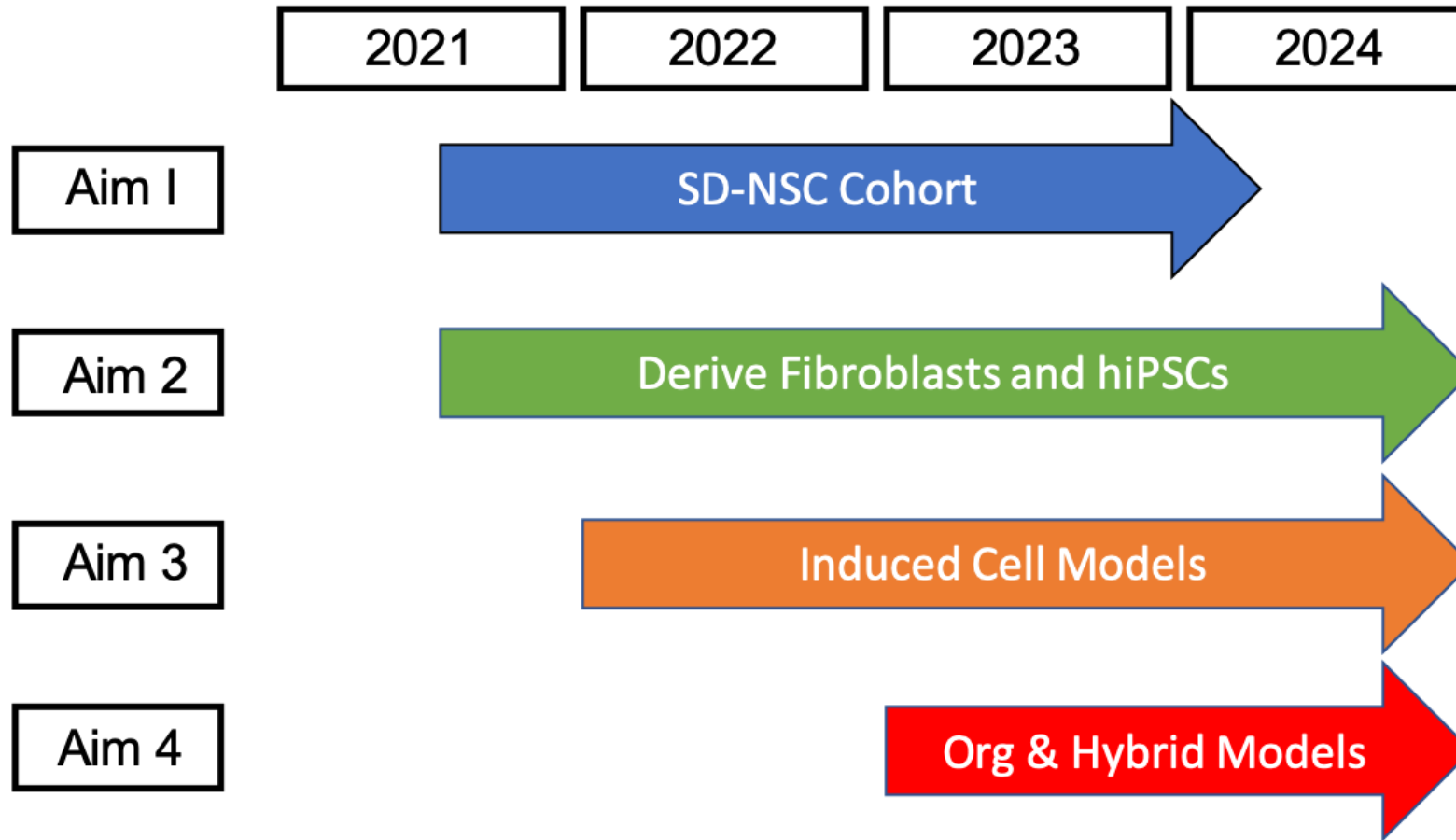
In development – HCMA Core partnering with key SD-NSC researchers (Rusty Gage)

Keep fingers crossed and stay tuned!

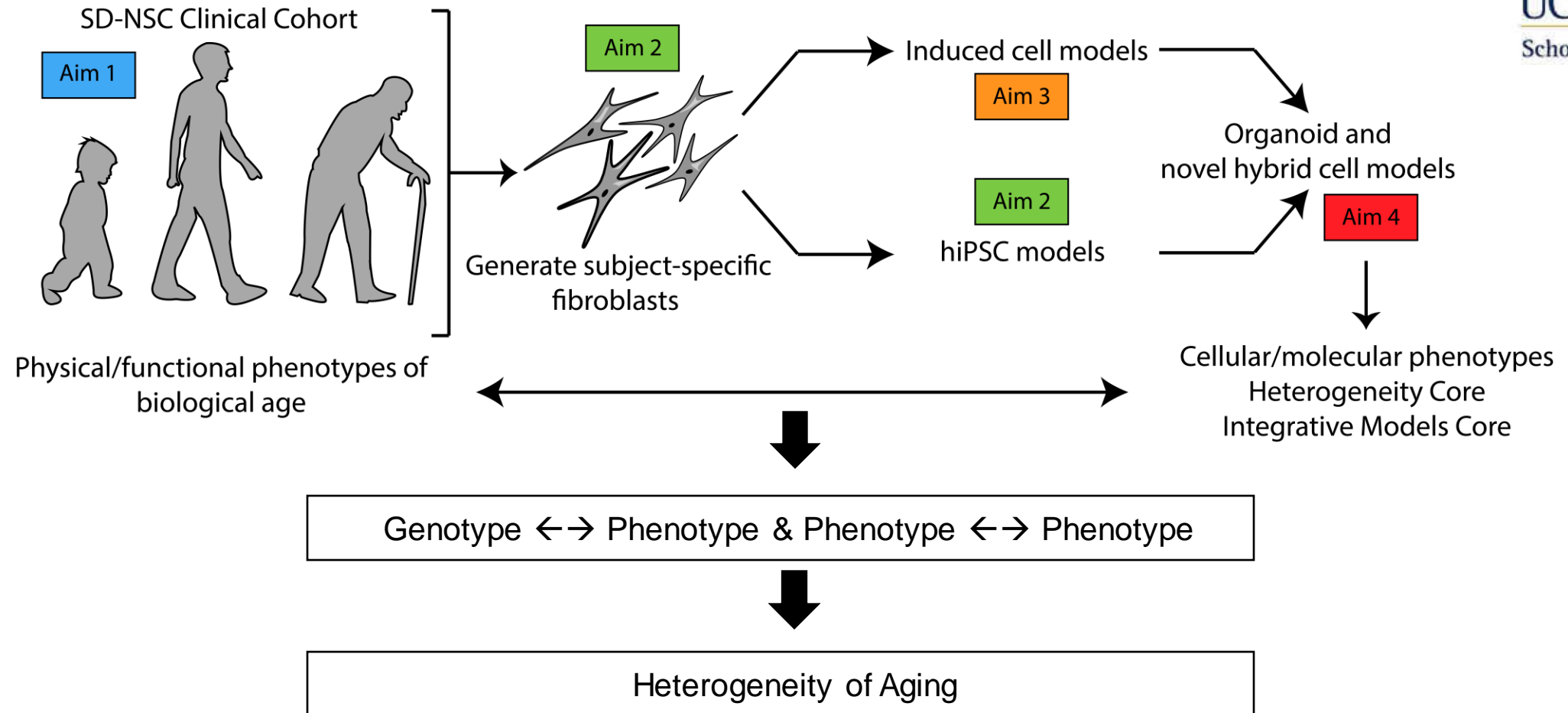
ESTIMATED TIMELINE FOR CELL RESOURCES AND MODELS



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SD-NSC COHORT POWERS IT ALL...





Q&A

MODULE 3 – HUMAN CELL MODELS OF AGING CORE

salk
Where cures begin.

Please type your questions in the chat

FACILITY EQUIPPED FOR CELL MODELING AND SUPPORT NEEDS

Overview of Facility, Equipment, and Resources

- 2500 sqft High Volume TC facility
- 13 hoods, 24 incubators
- Vapor-phase cryostorage
- Established 2007
- Newly renovated in 2012



FACILITY EQUIPPED FOR CELL MODELING AND SUPPORT NEEDS

Overview of Facility, Equipment, and Resources

The Mission: Lower the bar to access advance state of the art human cell based models

- Equipment (Live Imaging, Metabolism)
- Validated Media and Reagents
- Training and Project Support



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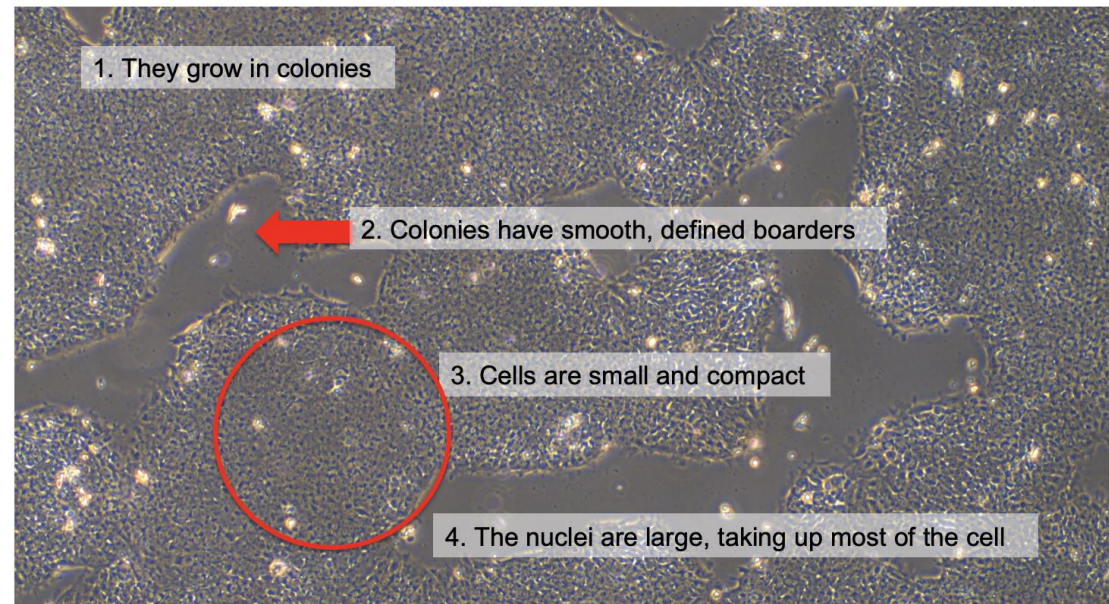


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