

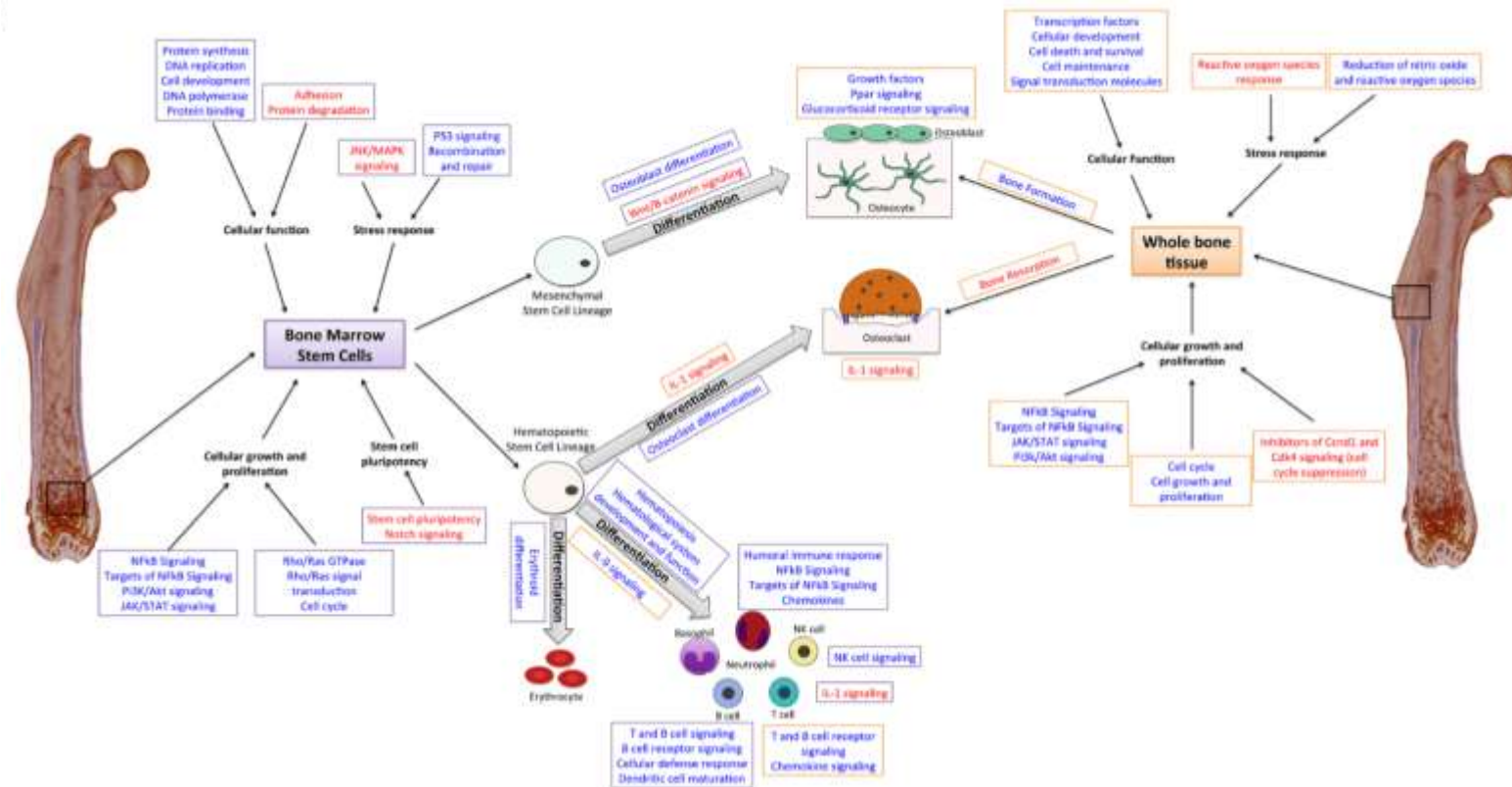
CDKN1a/p21 Plays Critical Role in Suppressing Stem Cell Regenerative Potential during Aging

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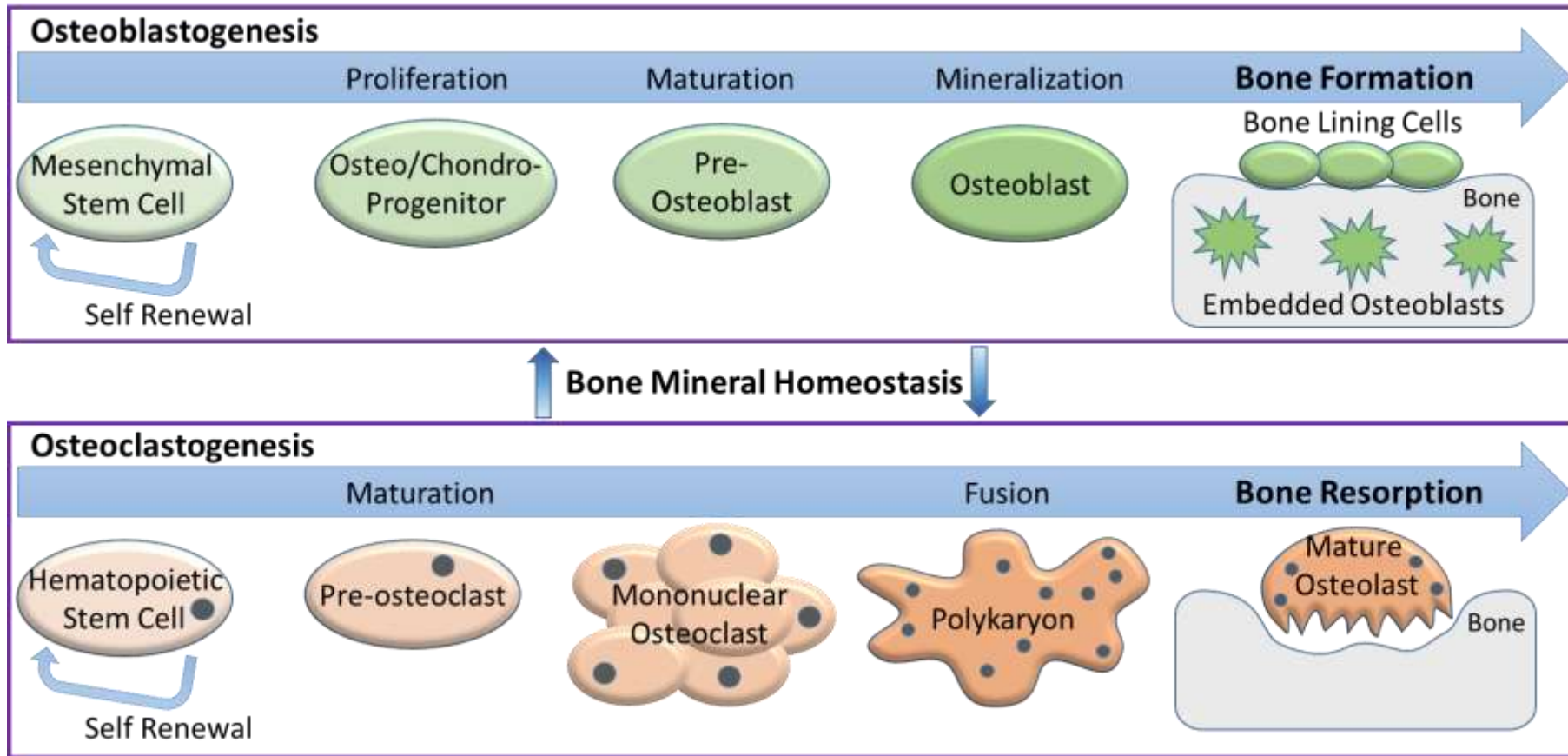


Bone in Spaceflight



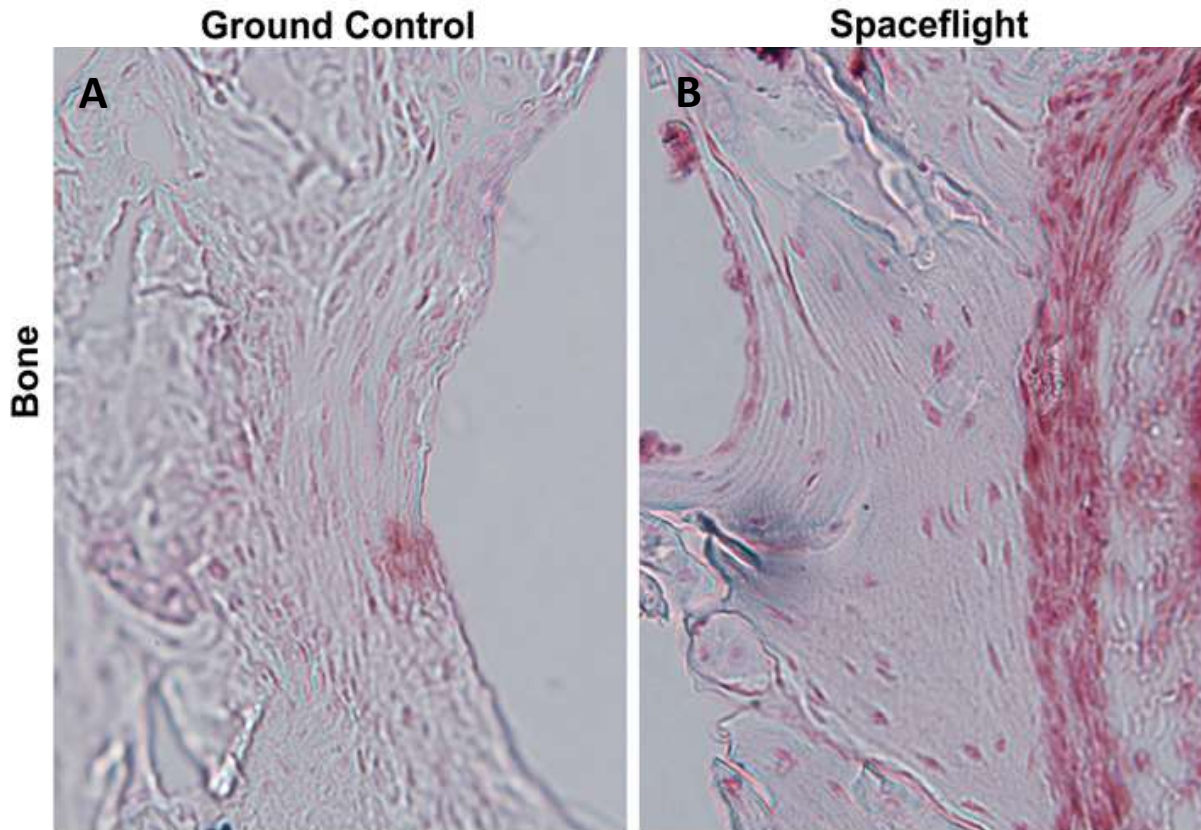
Spaceflight results in significant **down-regulation** of key genes required for mesenchymal and hematopoietic stem cell differentiation into terminally differentiated lineages.

Bone Regulation



Bone mineral homeostasis is a balance between bone formation by osteoblasts and bone resorption by osteoclasts.

CDKN1a/p21

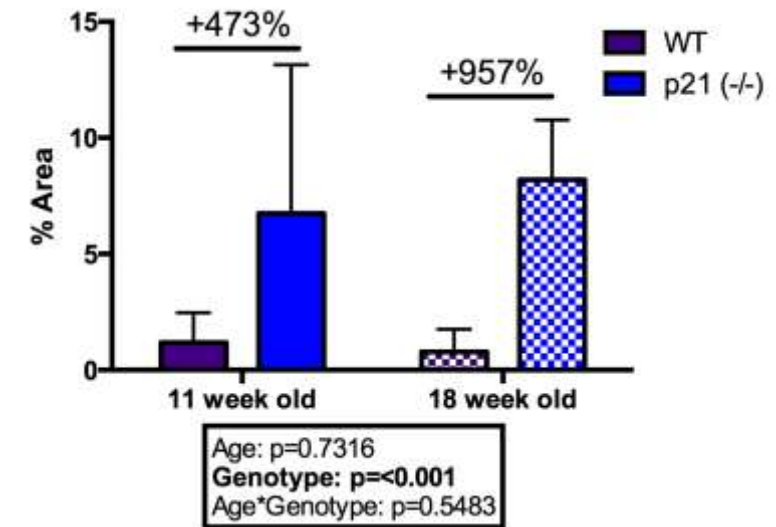
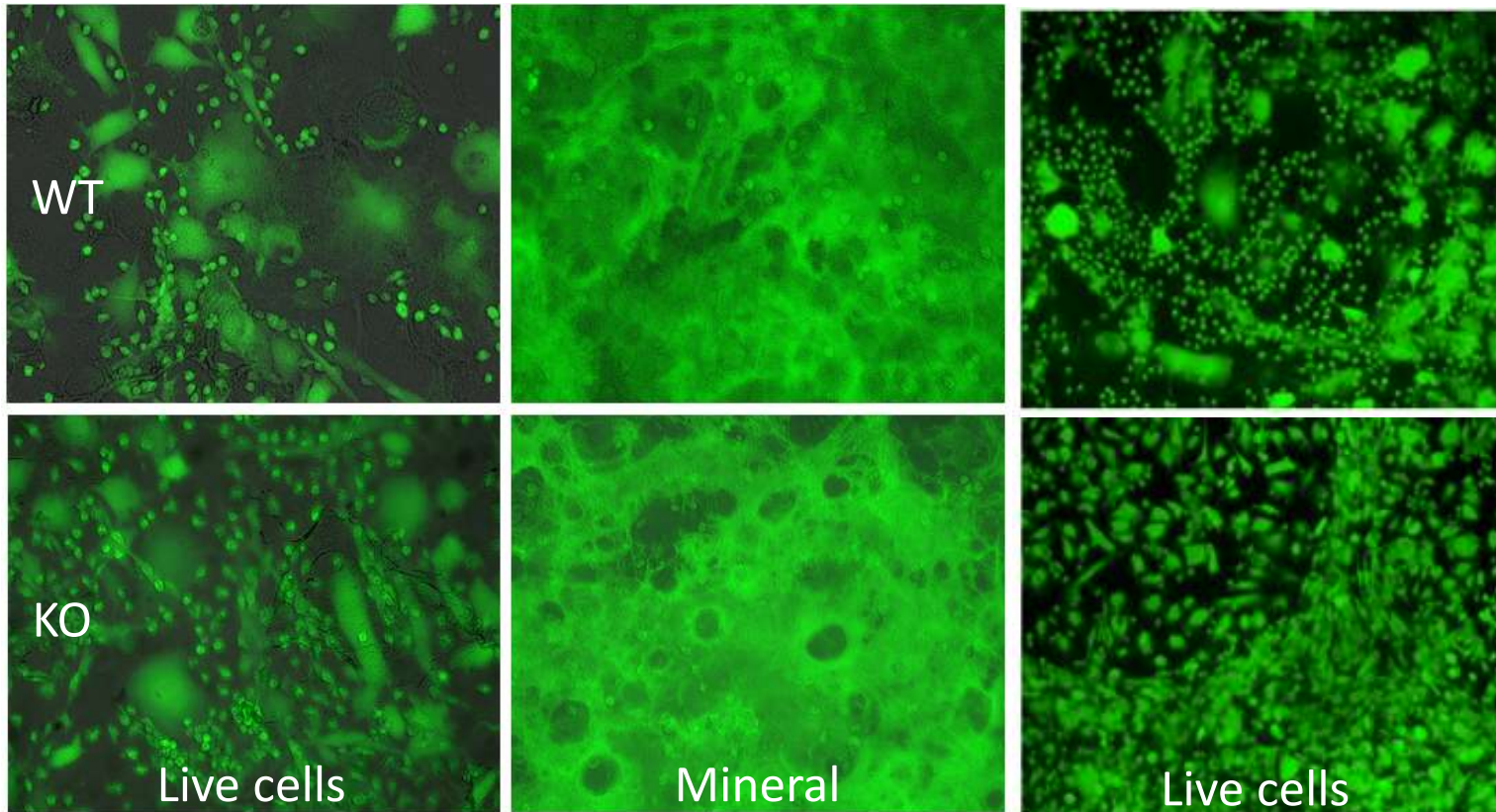


- Spaceflight mice showed increased CDKN1a/p21 in osteoprogenitor cells
- CDKN1a/p21 is a potent cell cycle arrest molecule
- CDKN1a/p21 knockout (KO) mice exhibit regenerative abilities similar to amphibians
- Ongoing studies will study the effect of age on proliferation and differentiation

Aging Comparison- WT vs KO

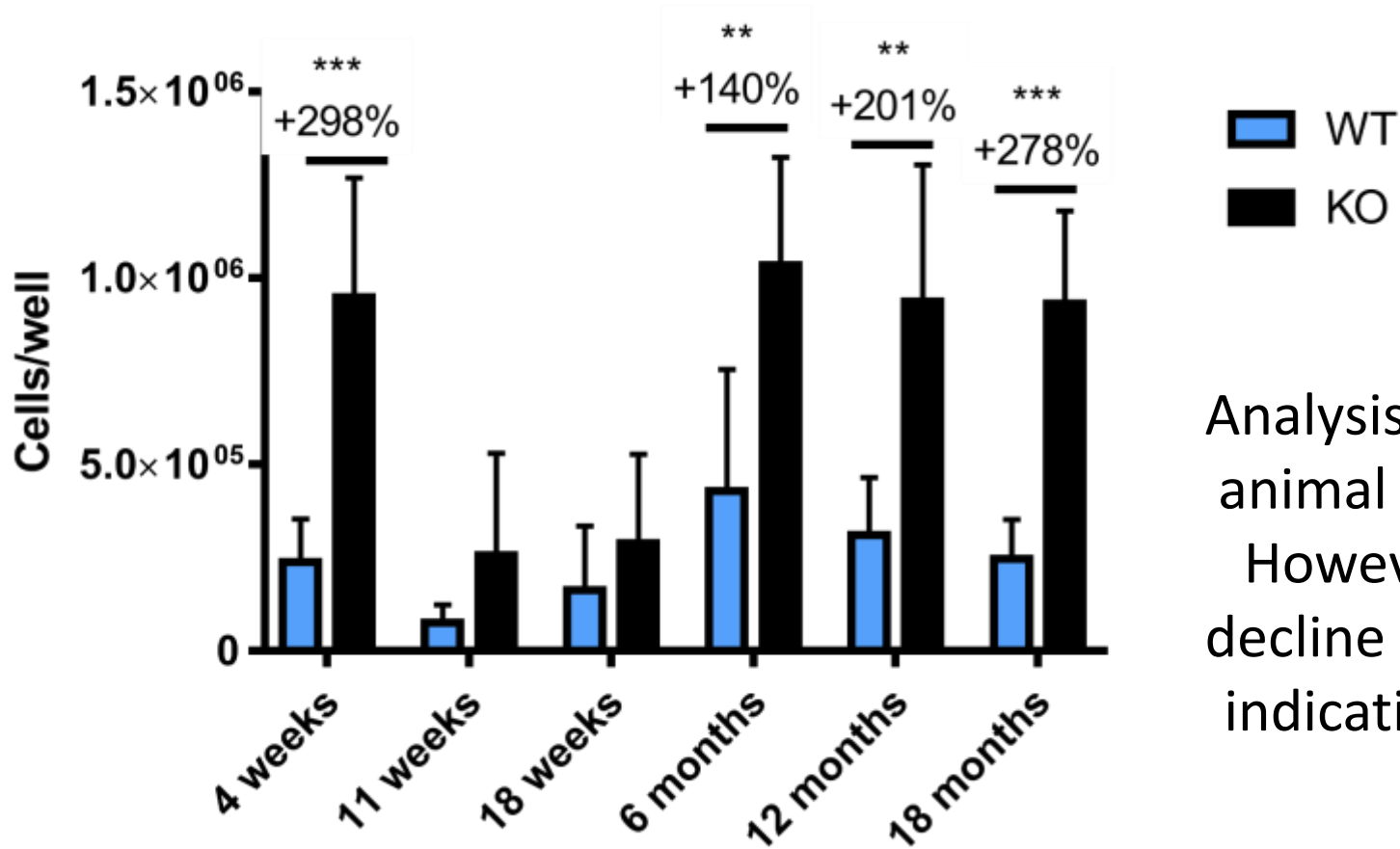
Juvenile Mice

Adult Mice



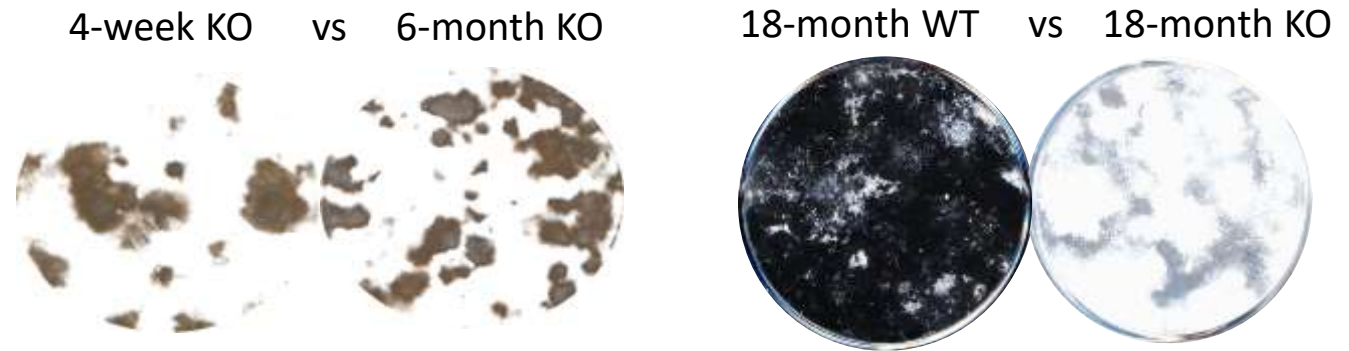
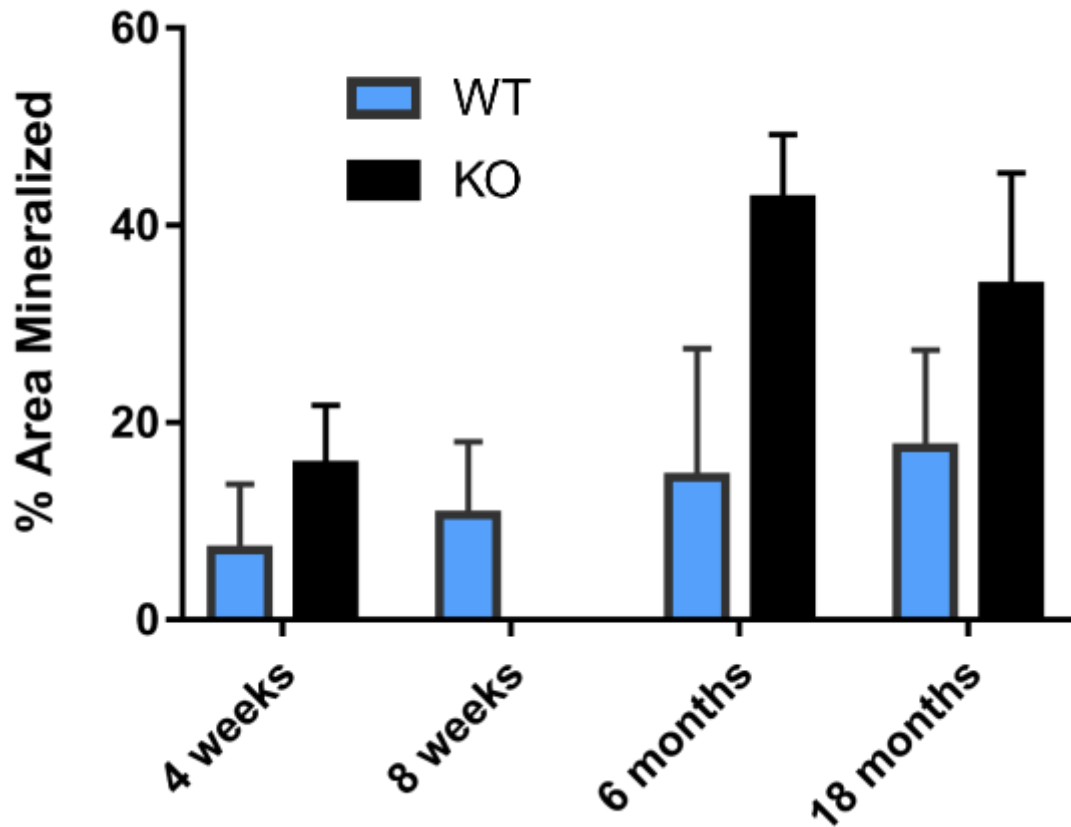
Calcein staining shows increased cell numbers and mineral in KO cultures compared to WT.

Aging Comparison- WT vs KO



Analysis of osteoblastic cultures indicate WT animal cell counts peak at 6 months of age. However, KO cells do not exhibit a similar decline in cell number past 6 months of age, indicating a **difference in proliferation and regenerative potential.**

Aging Comparison- WT vs KO

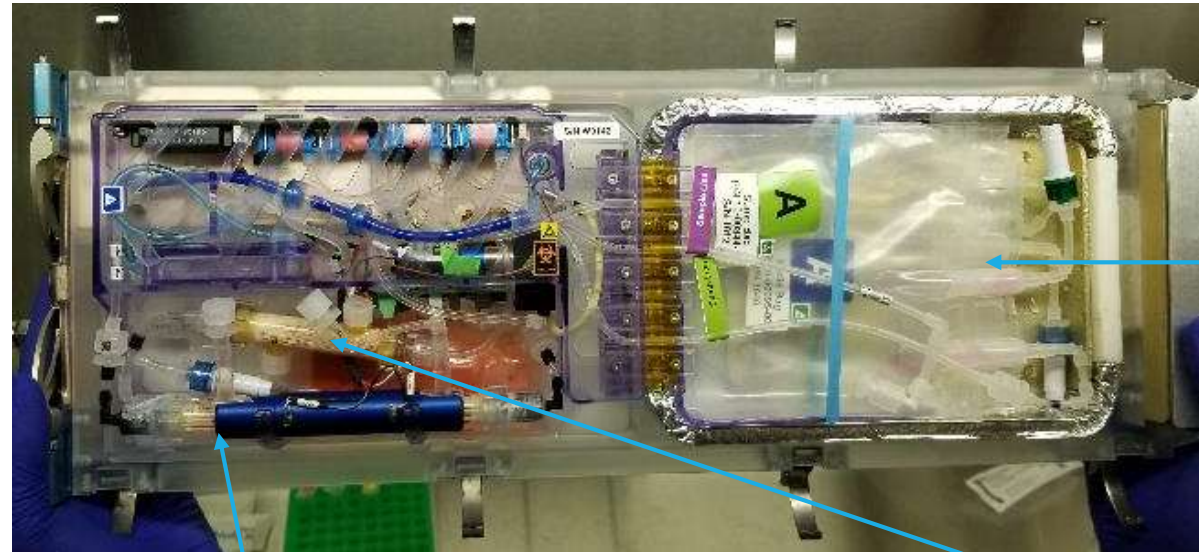


In addition to increased proliferation rates, deletion of CDKN1a/p21 in juvenile and adult mice resulted in **increased differentiation capacity** exhibited by increased formation of mineralized nodules.

CS-03 Flight Aims

1. To assess the in-vitro **proliferation, differentiation, and mineralization capacity** of BMSCs isolated from p21 KO and WT animals in microgravity versus 1g controls
2. To determine the **cellular mechanisms** associated with alterations in osteoprogenitor differentiation potential in p21 KO mice vs WT
3. To investigate the **signal transduction pathways** which are responsible for CDKN1a/p21 in microgravity and therefore inhibition of in vitro bone formation in space

Bioculture System



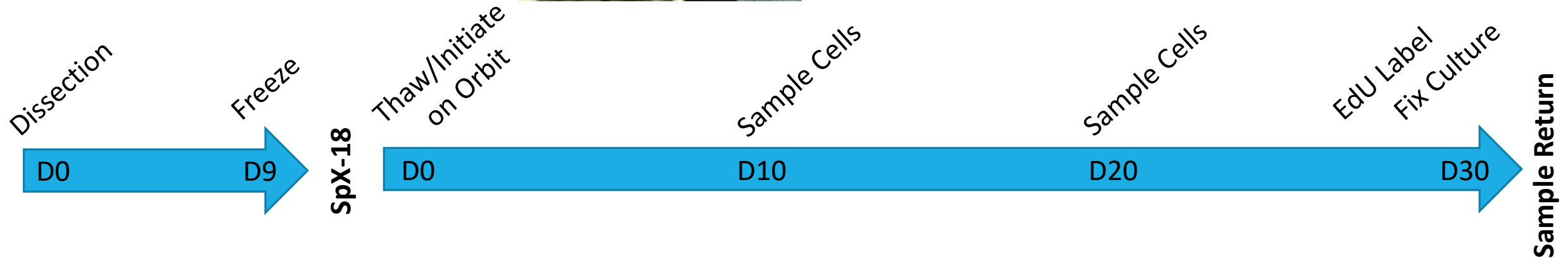
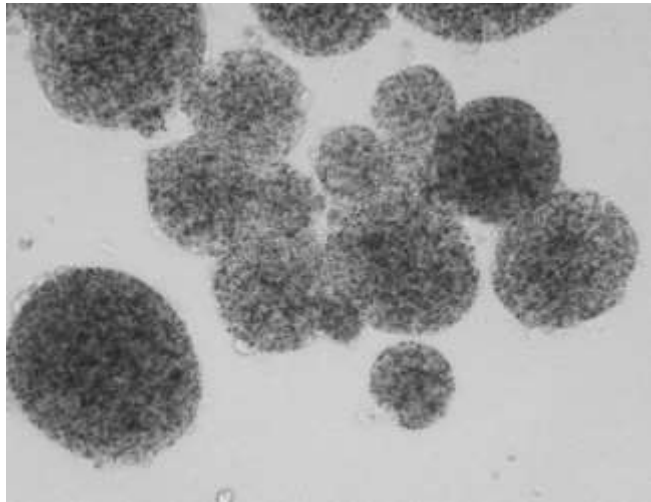
Media,
sump,
fixative, and
sample bags

Main Hollow-fiber Bioreactor

Secondary Growth Factor Bioreactor



Bioculture System- Osteoblastogenesis on Beads

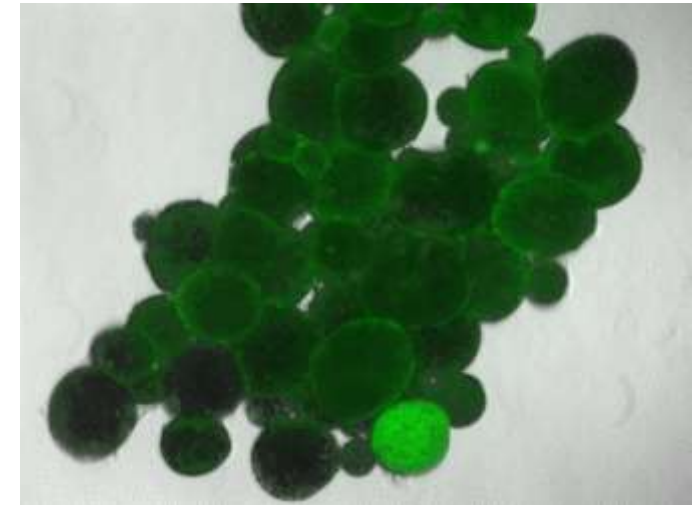
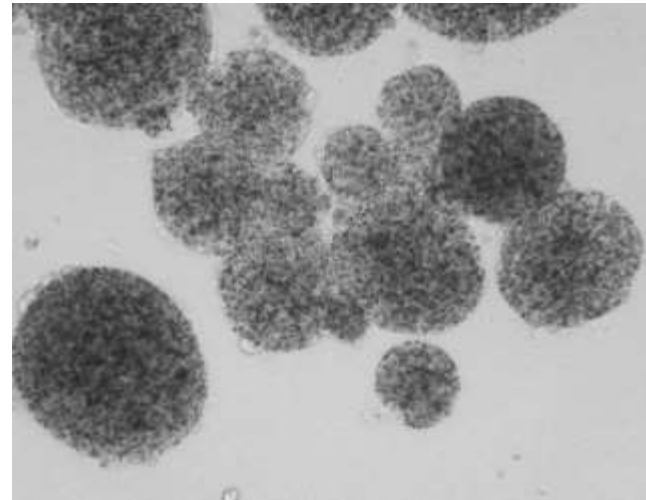
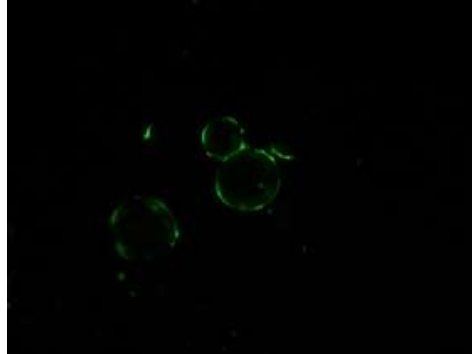
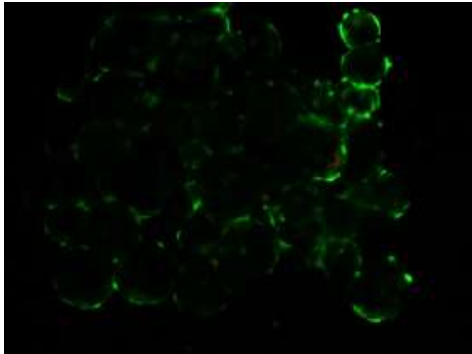


Osteoblastogenesis Using Gelatin Beads

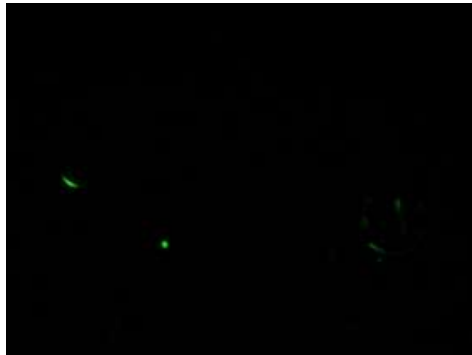
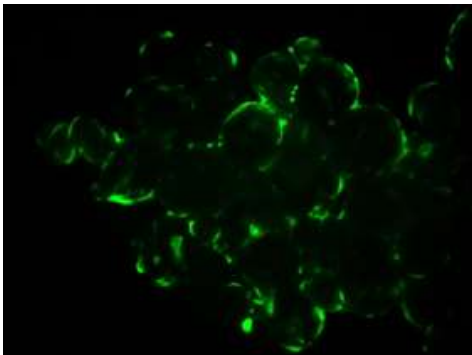
Knockout (KO)

Wildtype (WT)

Plate Controls

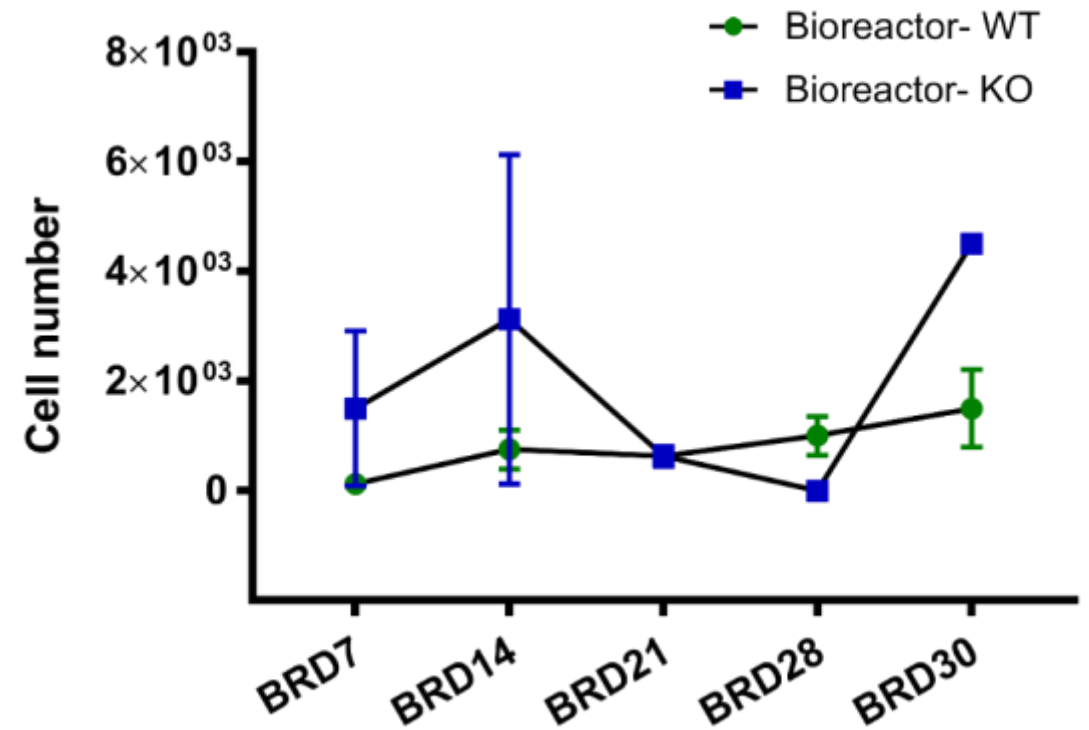
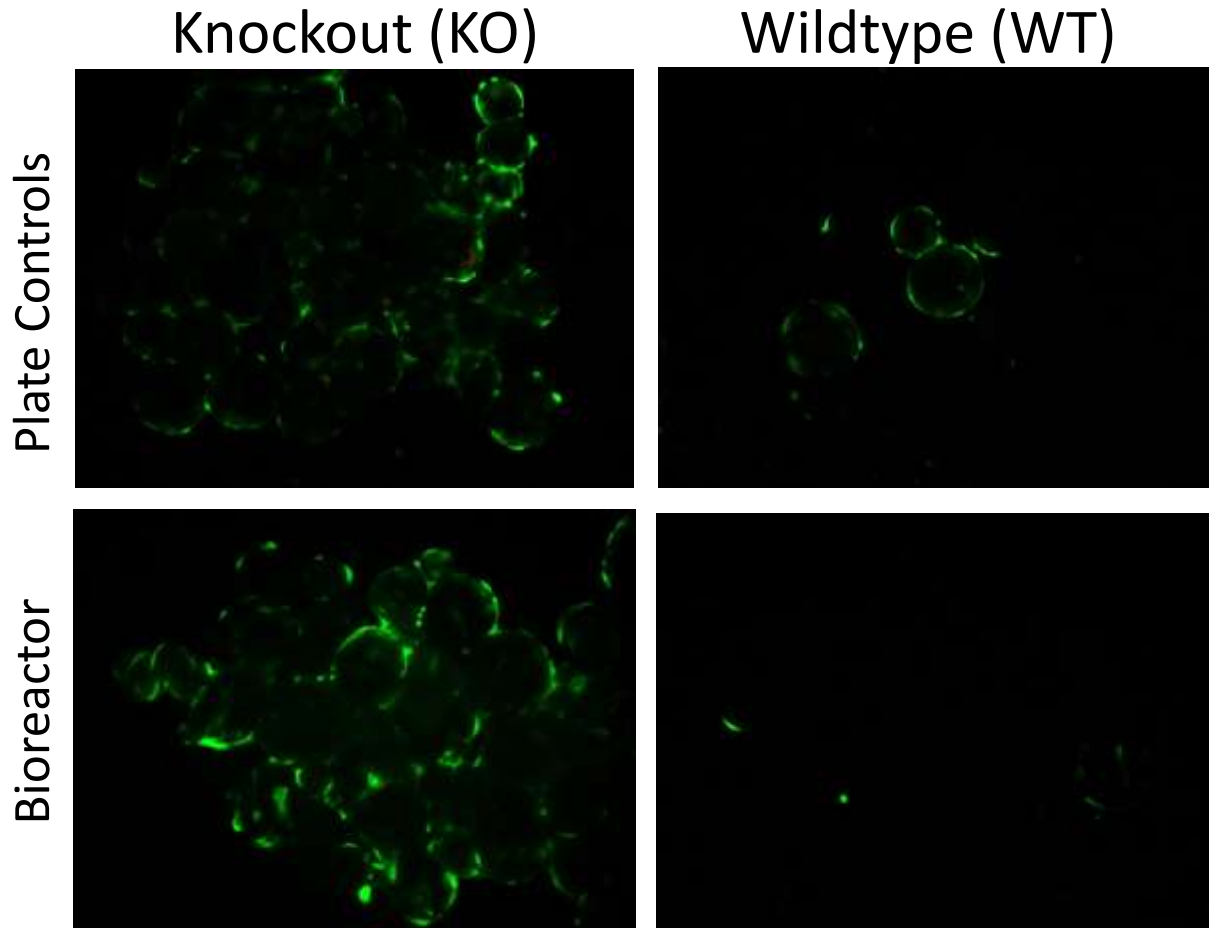


Bioreactor

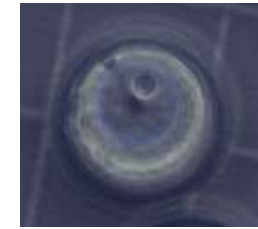


Global Eukaryotic Microcarriers
mineralized by cells as shown
by calcein stain (above)

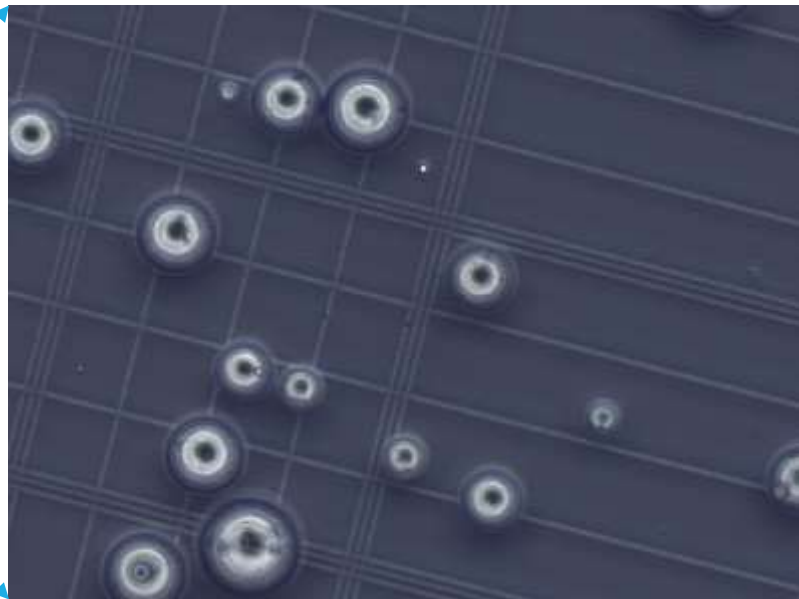
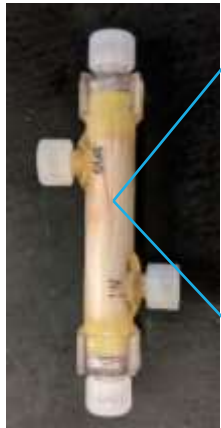
Osteoblastogenesis Using Gelatin Beads



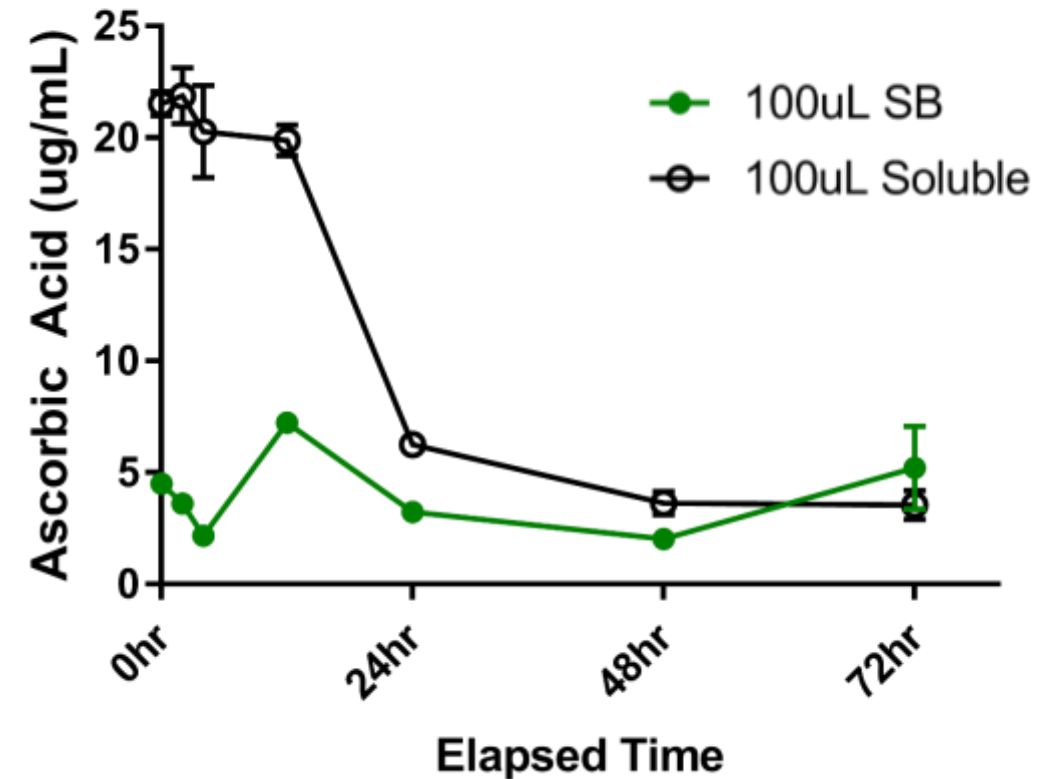
Growth Factor *StemBeads*



“That’s no moon”

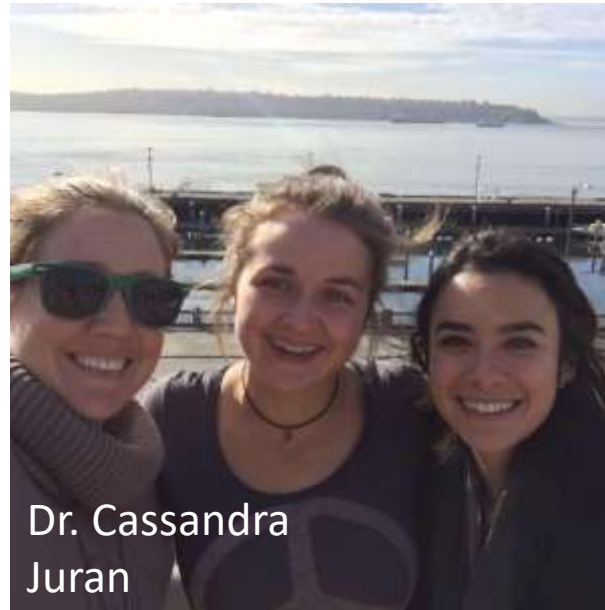


Ascorbic Acid Release Profile





Blue Marble Space
Institute of Science



Dr. Cassandra
Juran

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- Bioculture System Team
- StemCultures



Dr. Eduardo Almeida & Dr. Elizabeth Blaber

