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CENTER FOR DISRUPTIVE  
MUSCULOSKELETAL INNOVATIONS

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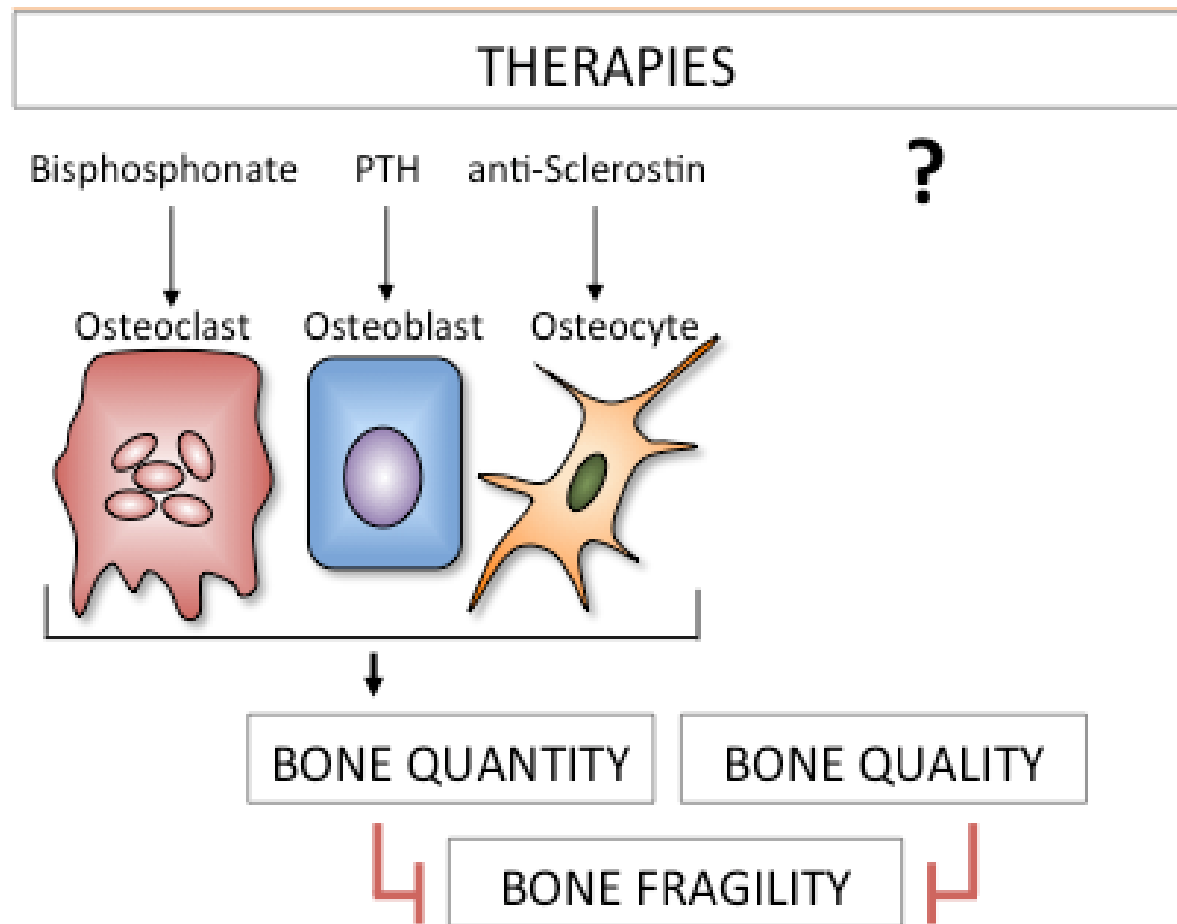
***Integrated in vivo and in vitro  
high-throughput analyses of  
osteocyte-mediated bone remodeling***

Tamara Alliston, Ph.D.  
UNIVERSITY OF CALIFORNIA SAN FRANCISCO

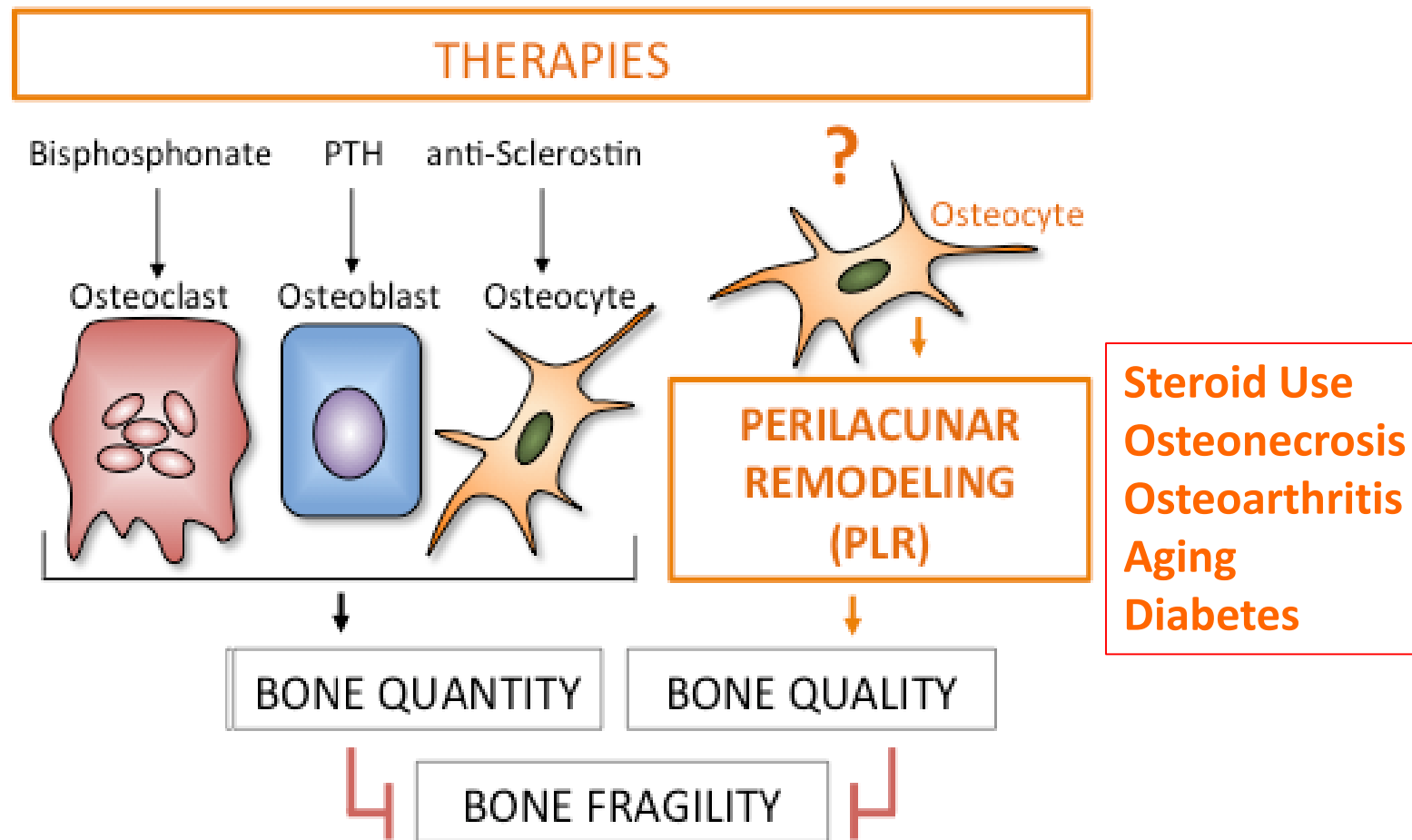
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[WWW.NSFCDMI.ORG](http://WWW.NSFCDMI.ORG)

# Clinical Imperative: Treat Bone Quality



# Clinical Need and Industrial Relevance



**Knowledge Gap:** role of PLR in bone health or disease & how to study it

## Project Aims

This project aims to develop a comprehensive approach to evaluate PLR in vivo and in vitro to advance the development of diagnostics and therapies to improve bone quality.

***Aim 1: Develop and validate in vitro measures of PLR function for high throughput screening.***

***- currently, there is no validated in vitro PLR assay***

***Aim 2: Establish the Osteocyte-Mediated Bone Remodeling ECM (OMBRE) Core.***

## ***OMBRE Protocols I-V*** 1

***I:*** Collagen Organization

***II:*** Lacunocanalicular Analysis

***III:*** PLR Gene Expression

***IV:*** In Vitro Functional pH Assay

***V:*** In Vitro PLR Reporter Assay



***High Throughput Screen*** 2

***CDMI Access to OMBRE Services through the UCSF Skeletal Biology and Biomechanics Core*** 3

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***High Throughput Screen***

***CDMI Access to OMBRE Services through the UCSF Skeletal Biology and Biomechanics Core***



# Core Center for Musculoskeletal Biology and Medicine



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## Skeletal Biology Core Services



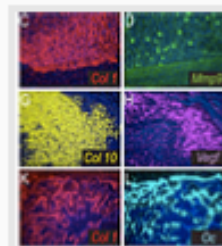
### Imaging and Histology Sub-Core

Provides imaging services for small animals and tissue specimens using computed X-ray tomography. Offers technical support in tissue extraction and processing for histology, as well as in histomorphometric analysis.



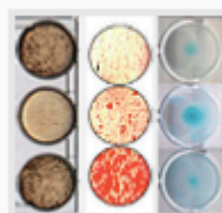
### Biomechanics Sub-Core

Offers resources and expertise in quantitatively evaluating the mechanical and material properties of skeletal tissue over a range of resolutions and scales.



### Molecular Biology Sub-Core

Provides expertise in the isolation of RNA and protein from skeletal tissue, the quantitative analysis of gene and protein expression using real-time qPCR and Western blotting, and the qualitative analysis of gene and protein expression through in situ hybridization and immunohistochemistry.



### Cell Culture Sub-Core

Maintains and supplies a collection of chondrocytic, osteoblastic, and osteoclastic cell lines and provides expertise in preparing primary cultures of murine bone marrow stromal cells and other cell populations. Offers technical support in the use of stains and enzymatic assays to assess bone-related properties in cultures.

Click [HERE](#) for a list of currently available cell lines.

OMBRE  
Core Services

UCSF Skeletal Biology  
Core Director:  
Tamara Alliston

Services Available  
via Recharge



# OMBRE Core Services



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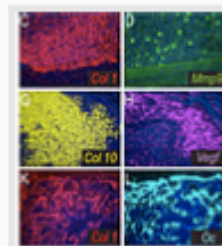
### Skeletal Biology Core Services



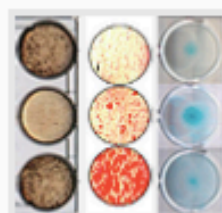
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**UCSF Skeletal Biology  
Core Director:  
Tamara Alliston**

**1 Progress Update**  
Recruitment of  
Cristal Yee, Ph.D.

Services Available  
via Recharge

**2 Progress Update**  
Preparing  
application for  
OMBRE Recharges  
(Feb 27, 2017)





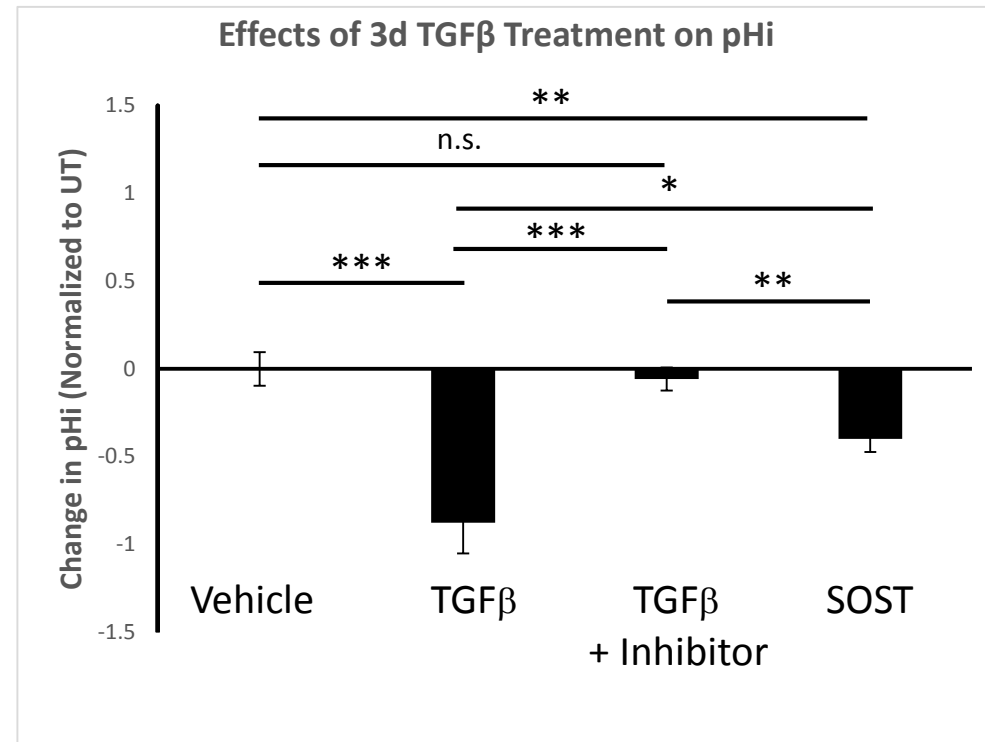
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## Progress Update

Developed & validated in vitro pH assay  
- pH regulation matches PLR regulation



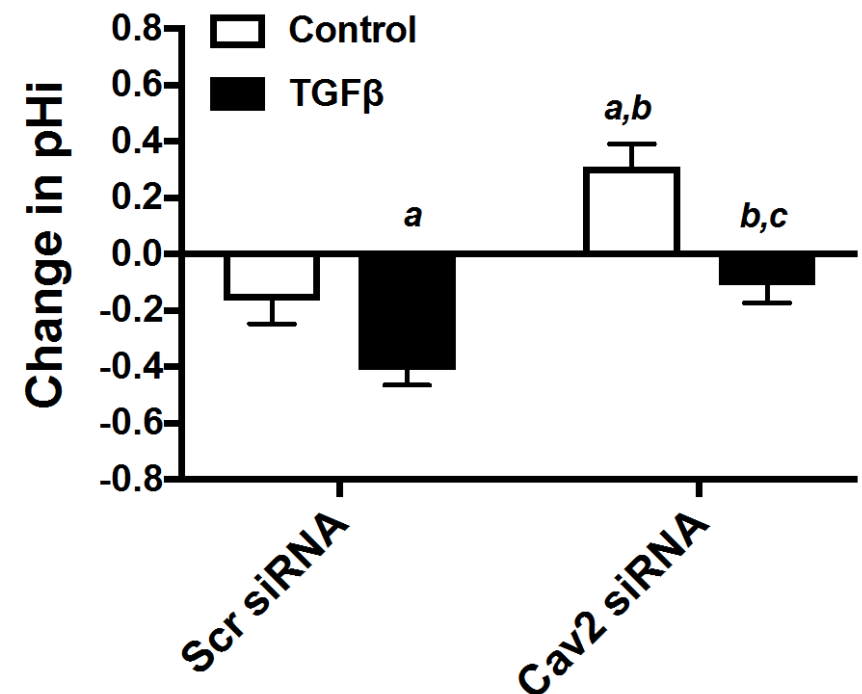
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### Progress Update

- Developed & validated in vitro pH assay
- pH regulation matches PLR regulation
  - sensitive in GOF and LOF assays



## OMBRE Protocols I-V

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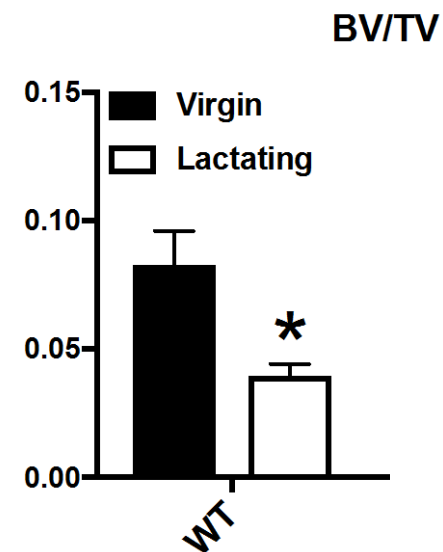
**V:** In Vitro PLR Reporter Assay

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## Progress Update

Harvested tissues for OMBRE validation

- virgin and lactating mice
- N  $\geq$  7 females per group
- microCT validation complete
- histology underway



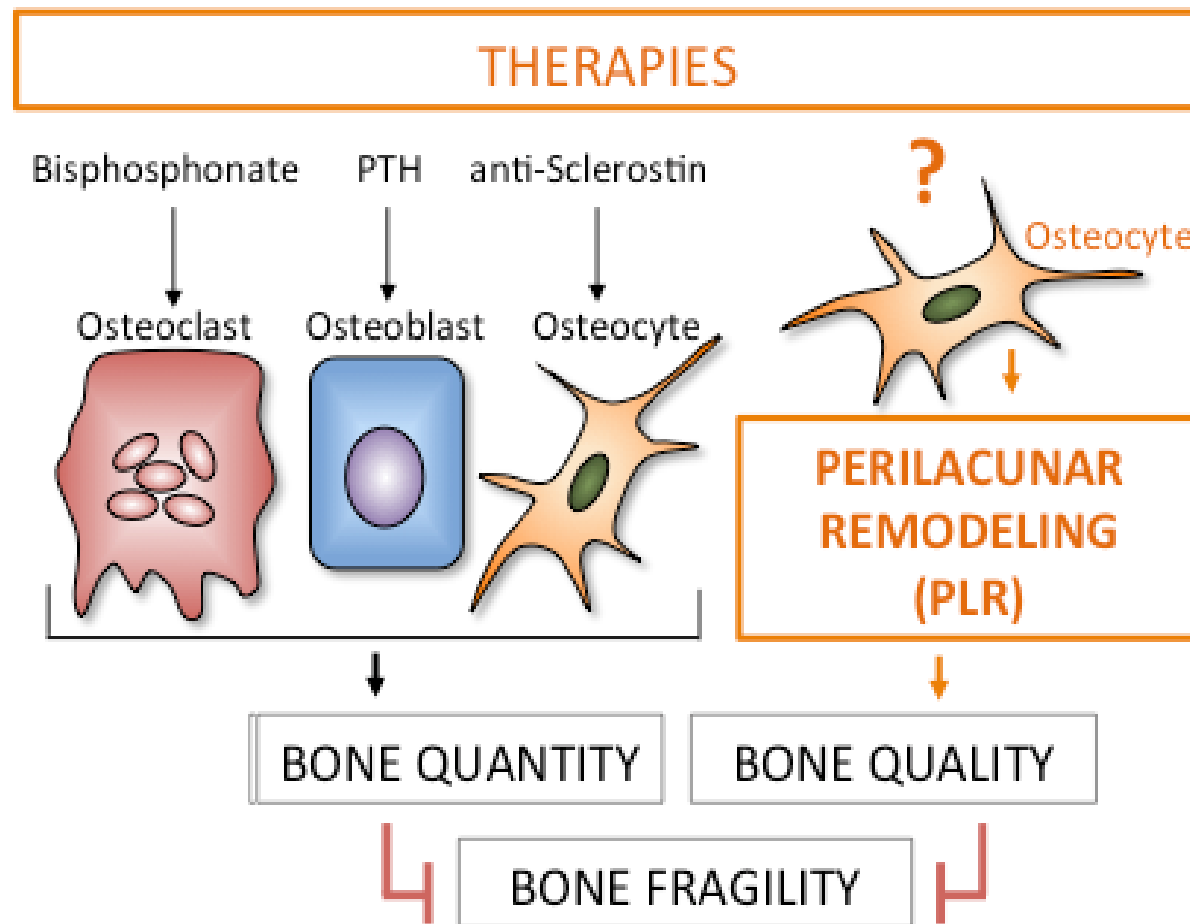
# Milestones

- Identify prototypical PLR-inducible gene for 1st in vitro PLR functional outcome
  - Dec 1, 2016
  - **STATUS:** MMP13, CatK, MMP14, MMP2, testing this month in SBB Core format
- Final protocol for OMBRE I: Collagen Organization
  - January 15, 2017
  - **STATUS:** Protocol complete, validating this quarter in lactating mouse bone
- Finish development of 2nd in vitro PLR functional outcome: intracellular pH assay
  - February 1, 2017
  - **STATUS:** Assay development and validation complete, writing protocol

# Clinical Need and Industrial Relevance

## Contributors

Justin Lopez  
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JJ Woo  
David Monteiro  
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Tristan Fowler  
Jackie Nguyen



Steroid Use  
Osteonecrosis  
Osteoarthritis  
Aging  
Diabetes