CDMI

CENTER FOR DISRUPTIVE MUSCULOSKELETAL INNOVATIONS

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

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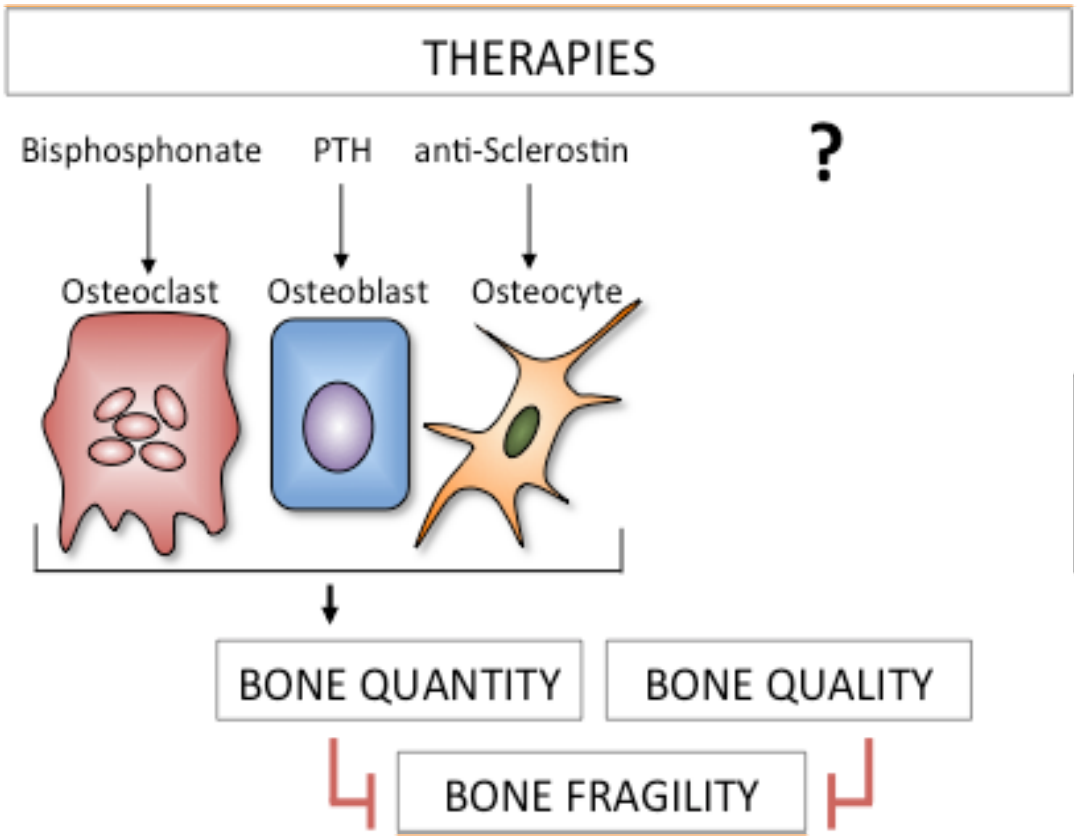
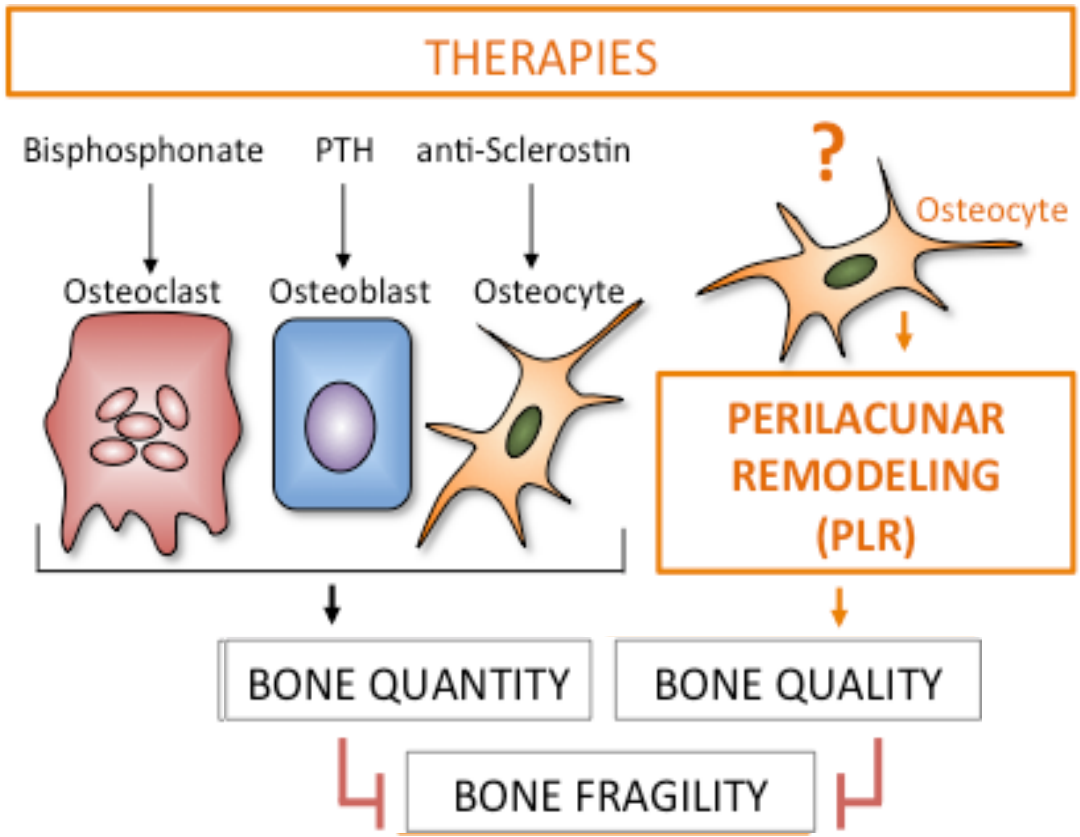
WWW.NSFCDMI.ORG

Clinical Imperative: Treat Bone Quality



Clinical Need and Industrial Relevance



Steroid Use
Osteonecrosis
Osteoarthritis
Aging
Diabetes

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.

Deliverables

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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

] 2 *High Throughput Screen*

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

OMBRE Core Services

UCSF Skeletal Biology Core Director:
Tamara Alliston

1 **Progress Update**
OMBRE Recharges Proposal Approved (March 22, 2017)

2 **Progress Update**
Working on updating website: CCMBM

3 **Progress Update**
UCSF, UC Davis, Industry interest for services

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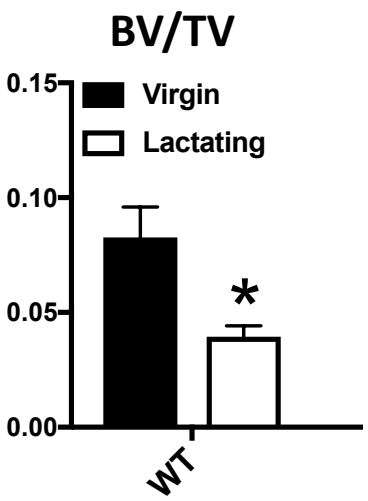
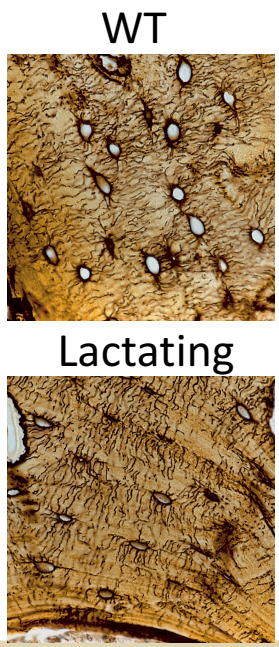
4

Progress Update

Harvested tissues for OMBRE validation

- virgin and lactating mice
- N ≥ 7 females per group
- microCT validation complete
- histology underway





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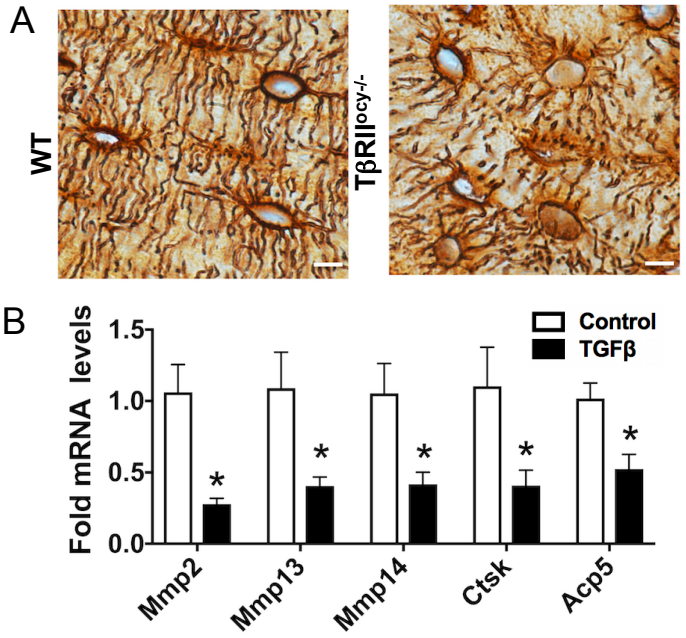
Progress Update

Harvested tissues for OMBRE validation

- Analysis of PLR Gene Expression
- PLR Gene Expression down regulated in TBRII$^{ocy-/-}$ mice



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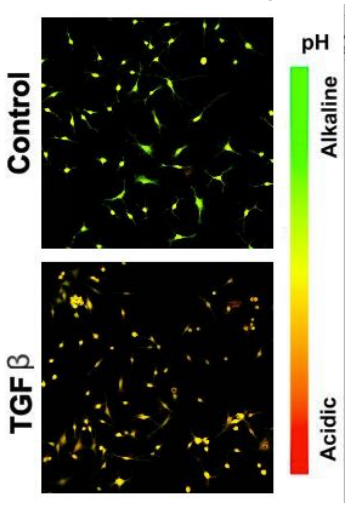
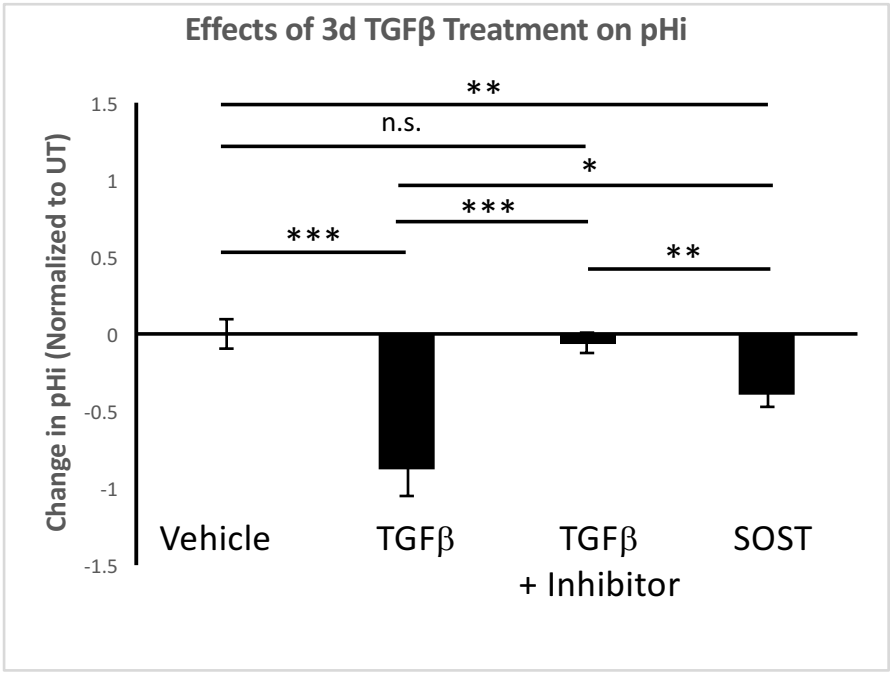


5 **Progress Update**

Developed & validated in vitro pH assay

- pH regulation matches PLR regulation
- performing viability assay in parallel with pH assay



Milestones

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In Vitro Assays

- Identify prototypical PLR-inducible gene for 1st in vitro PLR functional outcome
  - Dec 1, 2016
  - STATUS: New RNAseq data to be analyzed
- Finish cloning and sequence validation of novel PLR-report construct
  - April 1, 2017
  - STATUS: waiting for RNAseq data analysis to design desired clone
- Finish development of 2nd in vitro PLR functional outcome: intracellular pH assay
  - February 1, 2017
  - STATUS: Assay development and validation complete, writing protocol

In Vivo Assays

- Final protocol for OMBRE I: Collagen Organization
  - January 15, 2017
  - STATUS: Protocol complete, validated in lactating mouse bone
- Final protocol for OMBRE II: Canalicular Stain
  - March 1, 2017
  - STATUS: Protocol complete, proceed for quantitative analysis
- Finish protocol for OMBRE III: PLR Gene Expression
  - May 1, 2017
  - STATUS: Protocol complete, validated in TBRII$^{ocy-/-}$ mice

Clinical Need and Industrial Relevance

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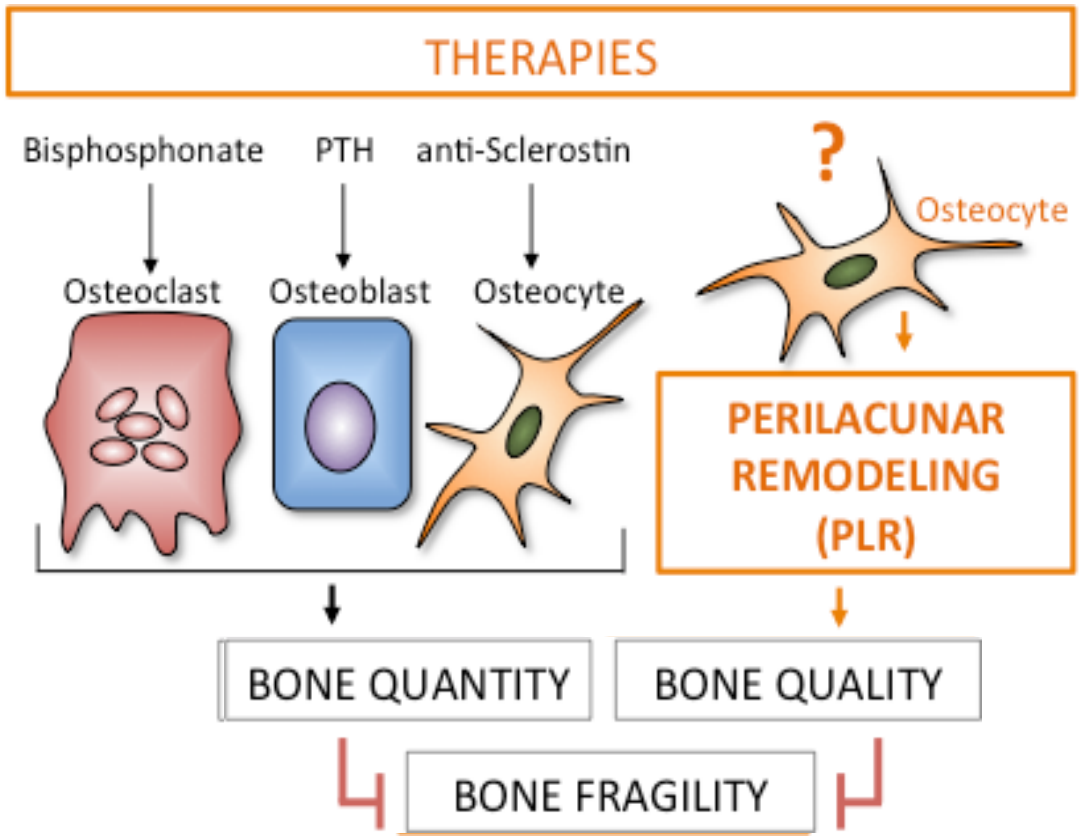

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