1. Introduction

Bone metastatic prostate cancer is incurable. Tumor progression in bone is driven by a network of cellular and molecular interactions, occurring both simultaneously and in parallel, that predispose regions of abnormal osteolysis and osteogenesis. TGFβ signaling is highly active in human prostate to bone metastases, yet because of its non-mutant effects it is difficult to fully define its effects on the tumor microenvironment. Thus, it is unknown if therapeutic inhibition would be efficacious.

Integration of computational modeling and biological systems allows for interrogation of conformational changes and signaling of TGFβ in response to multiple pathways, thereby predicting the outcome of therapy both in vitro and in vivo.

2. Hypothesis & Methods

Hypothesis: Using a discrete hybrid cellular automata model (HCA) combined with in vivo models, we can predict the efficacy of therapeutically targeting TGFβ in prostate cancer to bone metastases. To develop our HCA model, we parameterized partial differential equations with empirical and published data. In silico simulations of 250 days (n=127) were performed for five different levels of TGFβ inhibition (0-100%), supplied either pre- or post-metastatic seeding and were tested in vivo with an osteogenic and TGFβ-responsive model of bone metastatic prostate cancer (PaCa) using a TGFβ neutralizing antibody, 1D11.

3. Impact of in silico TGFβ inhibition on normal bone remodeling and tumor-induced bone formation

A Normal BMU

B PCa-BME

C TGFβ Inhibition increases normal bone formation and reduces tumor-induced osteogenesis. A: normal bone formation remodeling shown in snapshot (left) of HCA at Days 25 and 75. Live reads represent bone formation. B: TGFβ inhibition prior to tumor seeding (pre-treatment) inhibits tumor growth and bone formation. Left, images of treatment HCA model at Day 100 of 250 days; right, graphs represent tumor growth and bone formation at different TGFβ inhibitor doses, per-and-post-treatment.

4. In vivo TGFβ inhibition testing of in silico predictions of tumor growth

A In silico B In vivo

TGFβ inhibition does not exacerbate tumor-induced osteogenesis. A-C: Graphs illustrate impact of TGFβ inhibition on bone volume and osteoblast proliferation and apoptotic indices. D: Representative images (left) and quantitation (right) of microcomputed tomography (μCT) analysis; E-F: representative images and quantitation of in vivo bone volume to tissue volume (E) and total osteoblasts per mm bone(F) using Trichrome histological stain.

5. Impact of TGFβ inhibition on tumor-induced osteogenesis

A In silico B In vivo

TGFβ inhibition does not exacerbate tumor-induced osteogenesis. A-C: Representative images of the HCA model, PCa-BME prostate cancer-bone microenvironment; control (left) and with TGFβ inhibition(right). D: Graphs show in silico tumor burden proliferation and apoptotic indices, E: representative in vivo biomarkers (left) and quantitation (right) of SCD1 Beige mice inoculated with PaCa 60 days after 1D11 treatment. F: Osteoblast histological stain.

6. Impact of TGFβ inhibition on bone osteosclerosis in vivo and in silico

A B C

TGFβ inhibition reduces osteosclerosis. A-C: Representative graphs of total number of active osteoclasts (OCL), OCL maturation/fusion, and OCL activity. D: In vivo x-ray images of tumor-bearing tibia and quantitation of lesions per 2mm bone (right); E: images of TRAP-positive osteoclasts, graph represents total number of TRAP-positive OCLs per mm of bone in vivo.

7. Impact of TGFβ inhibition of Heterogenous Tumor using mCRPC patient data

A B C

TGFβ inhibition of heterogeneous tumor population identified outgrowth of resistant population. A: histological images of TGFβ ligand, receptor (TGFβR), and Smad signaling (pSMAD2) in human metastatic castration-resistant prostate cancer. Signal intensity was quantified using Definiens Tissue Studio (lower panel) and used for HCA model. B: Representative images (top) and quantitation (lower) panel of heterogeneous TGFβ expression in Patient 41458 with pre-treatment TGFβ inhibitor (right) at Day 100 of 250 day simulation.

8. Conclusions and Future Directions

- Integrated findings from our HCA and in vivo model demonstrate that TGFβ therapy would be beneficial for targeting bone metastatic prostate cancer when applied at early stages in tumor progression.
- Using the HCA computational model, we were able to examine changes in the tumor microenvironment at distinct time points and to identify the optimal therapeutic window for preventing tumor growth, without exacerbating tumor-induced osteogenesis.
- In combination with patient data, our HCA model can be utilized to identify emergence of tumor populations resistant to targeted therapy.

Future Directions:
- Our HCA computational model is broadly applicable to studying the tumor microenvironment of other cancer types. In the near future, the model will be used as a predictive tool for optimizing therapeutic pathways of bone metastatic prostate cancer.

9. Support

Supported by National Institutes of Health [RO1CA143304 (CCL)], the US Department of Defense [PC110460 (DAB)], and the American Cancer Society Postdoctoral Fellowship [PF-13-175-01-CSM (LC)]. The TGFβ inhibitor (1D11 antibody) and 13C control IgG antibody were supplied by Scott Lonning-Genzyme, Inc.