**Lay Description of Important Outcomes**

• Normal immune cells called macrophages can combine or merge together in bone to become osteoclasts, which normally remove bone so new bone can be formed. However, in breast cancer bone metastasis, osteoclast activity is increased by the tumor cells, resulting in bone loss, pain, and potentially fracture. Macrophage and subsequent osteoclast activity in bone is controlled by a protein named PU.1. PU.1 not only acts downstream of a cell surface protein called CSF1R, but PU.1 also controls the

expression of CSF1R on the cell surface of macrophages and osteoclasts. We have found that PU.1 also controls the function of macrophages and osteoclasts through its interaction with a family of proteins called BET proteins. We have found that targeting CSF1R or BET proteins pharmacologically in the laboratory are ways to inhibit macrophage and osteoclast activity. Since osteoclast inhibitors are one method to treat breast cancer bone metastasis, our goal in this proposal was to determine the effects of targeting CSF1R and BET proteins, alone and in combination, on breast cancer bone metastasis.

• We have found that combined inhibition of CSF1R and BET proteins is more effective that single agent therapy in reducing tumor growth. This finding that combined CSF1R and BET inhibition is more effective than monotherapy is extremely encouraging. One of our mouse models is one of triple negative breast cancer. CSF1R inhibitor resistance is a major clinical problem, most notably in triple negative breast cancer patients. This has limited the use of CSF1R inhibitors in these patients. Combined inhibition of both CSF1R and BET proteins may be one method to overcome CSF1R resistance, as well as expand

the use of BET inhibitors safely in breast cancer patients.