Internship project MMD2018

Cancer-induced osteolysis: near-physiological co-culture models to understand osteoclast-mediated bone resorption

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Background: Most cancer patients die as a consequence of their metastases. Many cancer types including lung, prostate and breast carcinomas metastasize to bone. About 70-80% of patients with advanced disease exhibit **bone metastasis resulting in bone lesions**, which can be osteoblastic, osteolytic or mixed. **Osteolytic bone lesions** not only can be fatal but also quickly worsen the patient quality of life because of loss of mobility, pathological fractures, nerve compression and debilitating cancer-induced bone pain. Existing systemic treatments are limited, thus, **broadening the repertoire of existing treatments against osteolytic bone metastasis is desirable**.

Research direction: Improving our **understanding** of cancer-induced osteolysis is crucial to improve **therapy effectiveness**. Osteolytic bone metastasis is caused by **cancer-induced hyperactivity of osteoclasts**, cells which resorb bone during tissue remodelling and (re)generation. We believe that understanding the dialogue between cancer cells and osteoclasts requires near physiological cell co-culture systems with breast cancer cells and osteoclasts that will enables us to unravel the regulation of metastasis-induced osteoclast-mediated bone resorption. To degrade bone, osteoclasts form a specialized **"sealing zone"** that warrants tight contact with the underlaying bone matrix and is composed by hundreds of small actin-rich cell protrusions, called podosomes, spatially arrange to surround the central bone resorption pit. We recently discovered that **phospholipase D (PLD) activity** is required to initiate podosome formation and extracellular matrix degradation in human dendritic cells and osteoclasts, but the underlying molecular mechanisms are still unclear. Small molecule inhibitors against PLD have decreased tumor growth and metastasis in mouse breast cancer, suggesting that they may be used to limit cancer cell growth and migration and perhaps also to reduce osteoclast-mediated osteolysis.

Aim: In this proposal, we will investigate the role of PLD activity in podosome-mediated bone resorption by osteoclasts and explore its potential as therapeutic target in our co-culture system. To this aim, we will develop co-cultures of human osteoclasts and breast cancer cells to study cancer cells-osteoclasts interplay in under semi-physiological conditions.

Methodologies. Preparation of human monocyte-derived osteoclasts (OCs); Complex transwell-based co-culture of human breast cancer cell lines and OCs; OC differentiation and functionality assays (e.g.: OsteoLyse™ Bone Resorption Assay Kit); Flow cytometry; Immunolabeling; Advanced fluorescence microscopy (i.e.: TIRF microscopy, super-resolution microscopy); Pharmacological inhibition of PLD signaling

Bibliography

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