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MOTIVATION

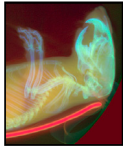
1. Conventional methods of studying preclinical models of metastasis involve serial sacrifice of large cohorts and are cumbersome.
2. *In vivo* imaging methods for estimating tumor burden with therapeutic efficacy testing are comparatively rapid and inherently more efficient.
3. *In vivo* optical imaging is being increasingly used in preclinical oncology to provide accurate and sensitive visualization of deep tissue tumors.
4. X-ray micro-CT provides high resolution anatomical context for characterizing preclinical bone metastasis models which is clinically relevant.

AIMS

1. To use *in vivo* bioluminescence imaging (BLI) to characterize the spread and growth of bone lesions in a model of PC-3 bone metastasis.
2. To use BLI-guided micro-CT to characterize the location and size of bone lesions.
3. To determine the feasibility of following bone metastases in the same animals by serial micro-CT images.
4. To investigate ways of enhancing metastasis visualization through direct volume rendering and multi-dimensional transfer functions.

INTRODUCTION

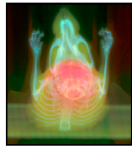
In vivo imaging in preclinical oncology



- *In vivo* imaging of animal tumor models is increasingly being used to accelerate efficacy evaluation in preclinical drug development.
- *In vivo* optical imaging such as bioluminescence (BLI), fluorescence and micro-PET provide enhanced detection with high sensitivity, but low resolution and limited anatomical context.

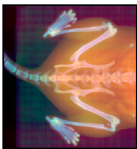
In vivo Micro-CT

- Micro-CT is increasingly being used to study mouse models of cancer *in vivo*.
- Micro-CT provides high resolution (50-100 μm, isotropic) images of normal and pathological mouse anatomy.



Combined BLI and Micro-CT

- *In vivo* BLI has the sensitivity to detect micro-metastases in a PC-3 bone metastasis model[†].
- Micro-CT is a clinically relevant approach to providing high resolution anatomical context in molecular imaging applications.



METHODS



Male ICR SCID mice were inoculated by intracardiac injection with 1x10⁶ PC-3.luc cells which constitutively express firefly luciferase.



Blue food dye was added to the cell suspension to provide visual verification of successful intracardiac injection.



Development of bone metastases was monitored by imaging with BLI under isoflurane anesthesia after IP injection of D-Luciferin.



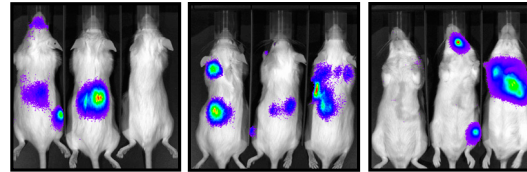
Results of BLI metastasis detection was used to guide serial weekly micro-CT of the same animals, enhancing the efficiency of micro-CT acquisition and analysis.



Organs and bones of tumored animals were harvested at necropsy for visual evaluation.

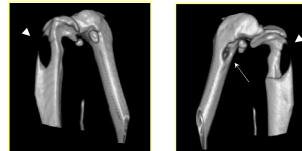
RESULTS and CONCLUSIONS

Bioluminescence Imaging for Detection of Metastases



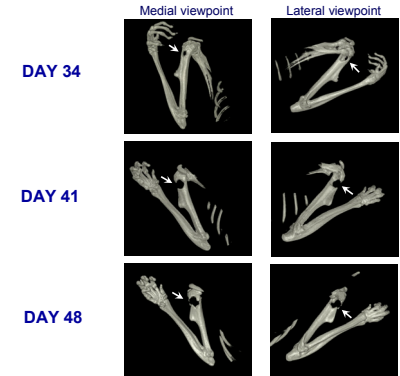
- Standard Bioluminescence Imaging was used to detect developing metastases.
- At necropsy, metastatic tumors were evident in heart, lung, and adrenal glands, as well as in the skull, mandible and long bones.
- 70% of injected animals developed bone metastases.

Volume Visualization of Bone Lesions



- Two views of a volume rendering of a large lesion on the proximal tibia (arrowheads). Another smaller lesion is evident on the femur (arrow).
- In micro-CT data, bone lesions appear as holes with size approximately proportional to the size of the metastatic tumor.

Time Course of Metastasis Development



- Metastasis in the right shoulder that was previously detected by Bioluminescence Imaging were followed by serial micro-CT (arrows).
- Bone destruction was clearly evident as tumor progressed.

Conclusions

- Bioluminescence imaging was able to detect and follow metastases *in vivo*.
- Metastatic tumors identified by Bioluminescence Imaging were detected and delineated as bone lesions by micro-CT *in vivo*.
- Changes in bone lesion size could be followed by serial micro-CT.
- Bone metastases were isolated in the micro-CT data and qualitatively visualized by robust volume rendering.

[†]Kalikin, LM et al. (2003), *Cancer Biology & Therapy*, 2, p17