Bone Metastasis Technology Platform - Establishing clinically relevant bone metastasis models for breast and prostate cancer and multiple myeloma

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Abstract number 3927,
Poster board number 4/20

Introduction

Bone metastases are a significant clinical problem in many major cancers, especially in breast and prostate cancer where 70-90% of advanced patients develop bone metastases. Myeloma bone disease is associated with similar clinical problems than bone metastases, including increased risk of fractures and bone pain that decrease the quality of life. No effective therapies are currently available for bone metastases, resulting in only 5% of bone metastatic patients being alive 5 years after the diagnosis. Bone metastases are therefore a high unmet medical need with a high demand for novel effective therapies.

Lack of appropriate preclinical bone metastasis models that would exhibit the same clinical features that are observed in bone metastatic patients has made it difficult to advance therapy development at early stages. Here, we describe a preclinical Bone Metastasis Technology Platform (BMTP)© that may solve the problem and substantially reduce the current >95% failure rate for oncology drugs in clinical trials, and especially costly failures in phase 3 due to poor efficacy.

Materials and Methods

This study describes the establishment of three preclinical bone metastasis models in BMTP:

- A breast cancer model using **4T1 mouse triple-negative breast cancer (TNBC)** cells in BALB/c mice
- A prostate cancer model using **RM-1 mouse castration resistant prostate cancer (CRPC)** cells in naïve and castrated C57BL/6 mice
- A **multiple myeloma (MM) bone disease model using RPMI-8226 human multiple myeloma** cells in immunodeficient NPG mice.

Cancer cells were inoculated intratibially into the bone marrow to model tumor growth in bone. Tumor growth was monitored by bioluminescence imaging (BLI), cancer-induced bone changes by X-ray imaging, and bone pain by Von Frey filaments (mechanical allodynia).

Results

Overview of the Bone Metastasis Technology Platform (BMTP)

Study start → **Randomization to groups, treatment start** → **Monitoring tumor burden, cancer-induced bone loss and bone pain during the study**

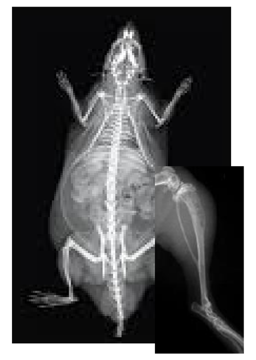
Cancer cell inoculation (Intratibial inoculation)

Direct injection of cancer cells to bone marrow, mimicking tumor growth in the bone microenvironment. Advantages of the intratibial inoculation include high tumor take rate and uniform tumor growth.

Tumor burden (BLI)

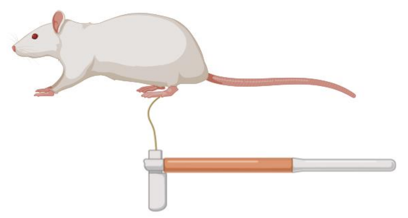
For determining tumor burden in models where tumor growth cannot be monitored by caliper. Advantages of BLI include the possibility for detection and quantitation of metastases.

Cancer-induced bone loss (X-ray imaging and scoring)



Quick and easy method for bone loss detection and monitoring during a study. Provides information on the extend of bone loss and/or drug associated bone effects.

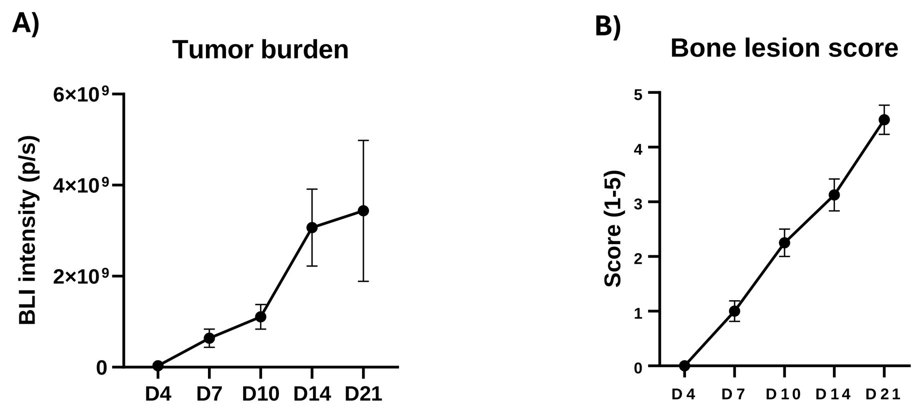
Bone pain (Mechanical allodynia by von Frey filaments)



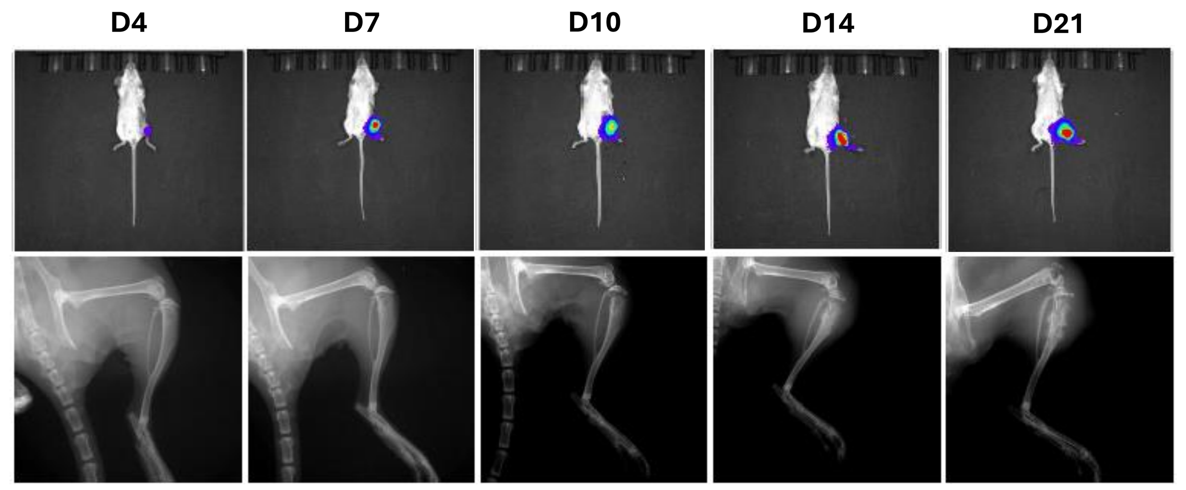
Non-invasive method for understanding extend of bone pain. Measurements can be performed multiple times during the study.

TNBC bone metastasis model

SUMMARY: In the 4T1 TNBC model, 100% of the mice had bone metastases at day 4, and maximum study duration was 21 days. Osteolytic bone lesions were clearly observed and bone pain was detected at day 7.



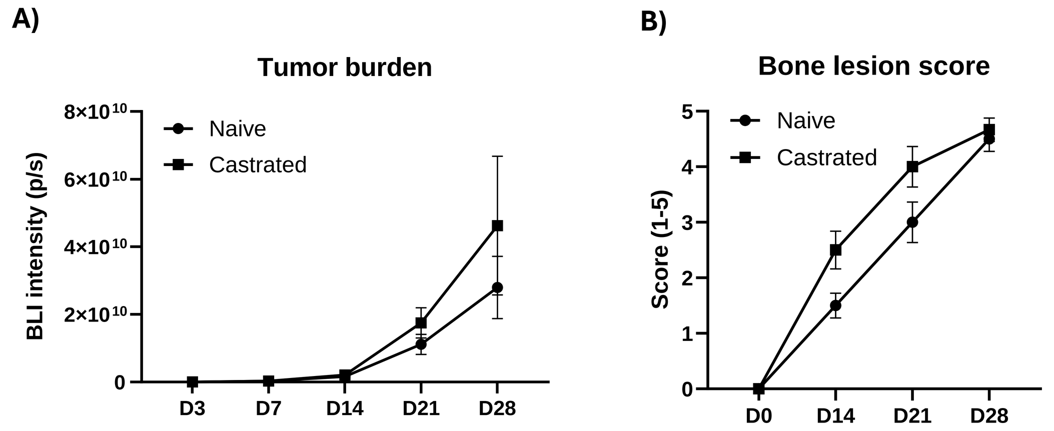
Development of tumor burden as determined by BLI (A) and bone lesions as determined by X-ray (B) during the study. Bone lesions were scored from 1 to 5 by visual estimation of the X-ray images.



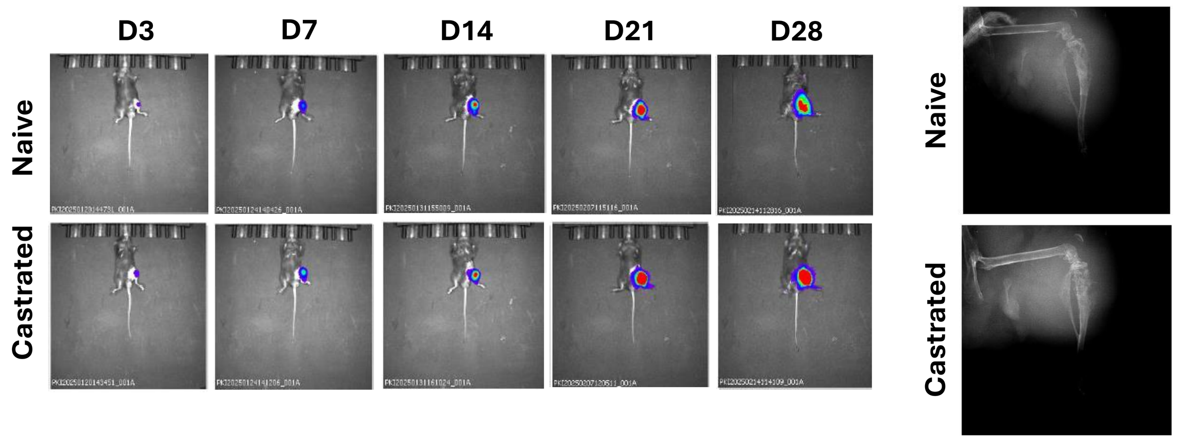
Disease development in a representative mouse. Upper row: Tumor burden detection by BLI during the study. Tumors were detected 4 days after the 4T1 cell inoculation. Lower row: Tumor-induced bone loss (bone lesions) visualized by X-ray imaging. Bone lesions were observed at 7 days after the 4T1 cell inoculation, and extensive bone loss was observed at the end of the study in all mice.

CRPC bone metastasis model

SUMMARY: In the RM-1 prostate cancer model, 100% of the mice had bone metastases at day 7, and maximum study duration was 28 days. Bone pain was observed at day 7, and osteolytic-mixed bone metastases were visible at day 14. Tumors grew and bone lesions formed similarly in naïve and castrated mice.



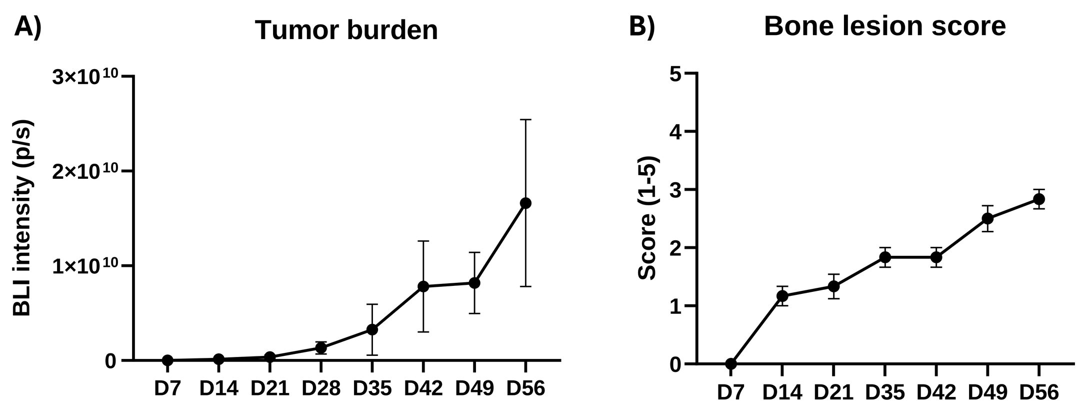
Development of tumor burden as determined by BLI (A) and bone lesions as determined by X-ray (B) during the study in naïve and castrated mice. Bone lesions were scored from 1 to 5 by visual estimation of the X-ray images.



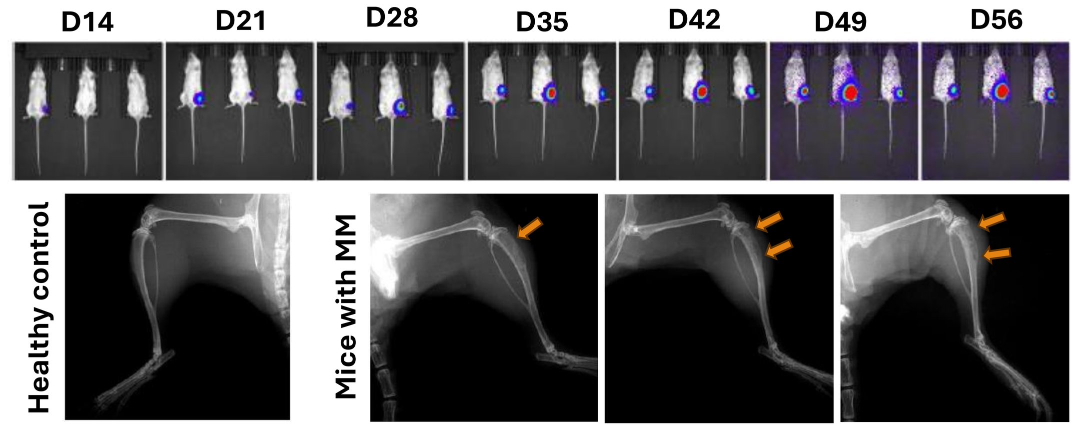
Disease development in a representative naïve and castrated mouse. On the left: Tumor burden detection by BLI during the study. Tumors were detected 3 days after the RM-1 cell inoculation. On the right: Tumor-induced bone loss (bone lesions) visualized by X-ray imaging. Bone lesions were observed at 14 days after the RM-1 cell inoculation, and extensive bone loss was observed at the end of the study in all mice.

MM bone disease model

SUMMARY: In the RPMI 8226 multiple myeloma model, 100% tumor take rate was detected at day 7. Osteolytic bone metastases were visible at day 21, and maximum study duration was 56 days.



Development of tumor burden as determined by BLI (A) and bone lesions as determined by X-ray (B) during the study. Bone lesions were scored from 1 to 5 by visual estimation of the X-ray images.



Disease development in representative mice. Upper row: Tumor burden detection by BLI during the study. Tumors were detected 7 days after RPMI-8226 cell inoculation. Lower row: Tumor-induced bone loss (bone lesions) visualized by X-ray imaging. Bone lesions were observed at 14 days after RPMI-8226 cell inoculation, and extensive bone loss was observed at the end of the study in all mice. Orange arrows indicate osteolytic bone loss.

Conclusions

We have established a clinically relevant Bone Metastasis Technology Platform (BMTP) that currently includes preclinical bone metastasis models for TNBC, CRPC and MM.

In the preclinical models established in BMTP, tumor burden is monitored by BLI, the type and extent of cancer-induced bone loss is visualized by X-ray imaging, and bone pain is analyzed to provide a clinically relevant readout about the quality of life.

The established models have clinical features similar to those observed in bone metastatic patients, including fast tumor growth and cancer-induced bone changes that lead to increased risk of fractures and bone pain, significantly decreasing the quality of life of bone metastatic patients.

We conclude that BMTP is a clinically relevant translational tool for evaluating efficacy of cancer therapies on bone metastasizing cancers, and it can be utilized to significantly reduce the current high failure rates of oncology drugs in clinical trials.

References

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