Castration-Resistant Prostate Cancer Bone Metastasis Model to Assess New Therapeutics

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Introduction

The role of androgens in the regulation of growth of primary prostate cancer is well established but less recognized in the context of advanced bone metastatic disease, which is the cause of high morbidity in patients. At advanced stage, prostate cancer cells typically lose their dependence on endogenous androgens, leading to development of metastatic castration-resistant prostate cancer (mCRPC).

The aim of the study was to establish a novel mCRPC model resembling important clinical aspects of bone metastatic disease that could be used for evaluation of efficacy of new therapies.

Materials and Methods

NOD-Scid male mice (Janvier) aged 5-6 weeks were divided into three study groups (n=10). Mice in two study groups were castrated either before or four weeks after cancer cell inoculation. The mice in one study group were left intact. All mice received an intratibial injection of 2x10⁶ VCaP human prostate cancer cells (ATCC) originally derived from vertebral metastatic site. Tumor growth was followed biweekly for 16 weeks by serum PSA measurements (R&D Systems) and X-ray imaging (Faxitron) of tumor induced bone changes (bone lesions), and ex vivo micro-computed tomography imaging (µCT, Bruker) of tibia. At endpoint, androgen-dependent organs were weighed and tumor-bearing tibias were subjected to histological evaluation and immunohistochemical staining of androgen receptor (AR; SP107) and AR splice variant 7 (AR-V7, both from Nordic BioSite).

Serum PSA

![Graph A](image1.png)

![Graph B](image2.png)

FIGURE 1. A) Prostate specific antigen (PSA) serum levels (ng/ml, mean ± SEM) measured during the study. Castration before cancer cell inoculation reduced PSA levels compared to intact mice (p<0.01 **). B) PSA values (ng/ml) from individual mice are presented.

X-ray and µCT analysis

![Graph A](image3.png)

![Graph B](image4.png)

FIGURE 2. A) Bone lesion area (mm², mean ± SEM) quantified from the X-ray images. No statistically significant differences were observed (p>0.250). B) Examples from X-ray images at endpoint. C) Examples from 3D reconstructions of tibia bones by µCT imaging showing changes in cortical bone.

Summary

- A tumor take rate of 60%, 40% and 50% was observed in the intact mice and in the mice castrated one week before and four weeks after VCaP cell inoculation, respectively. Tumor take rate was determined based on evaluation of PSA, bone lesions by X-ray imaging, and histological findings.
- The PSA levels were higher in the intact mice compared to the mice castrated before cancer cell inoculation.
- VCaP cells induced osteoblastic or mixed bone reaction and the lesions were similar in intact and castrated mice.
- Castration upregulated AR and AR-V7 expression in the tumor cells.
- Weight of androgen dependent organs was lower in the castrated mice compared to the intact mice.

Conclusions

A model mimicking important clinical aspects of castration-resistant prostate cancer bone metastases was established. Tumor take and growth rates indicated that early phases of tumor development into the bone is androgen regulated whereas tumor growth at later stage relies on the bone microenvironment for growth support. The results highlight the significance of the tumor microenvironment in establishing clinically relevant preclinical models for drug development.

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