Introduction

Immunotherapy for prostate cancer has recently emerged as an attractive treatment strategy. Yet, preclinical models where relationship between inflammation, stroma, tumor cells and prostate cancer progression can be studied are limited. In the present animal models, mainly human cancer cells are used in immunocompromised animals and interaction between the immune system and cancer cannot be monitored in these models due to the lack of active immune system.

Models with normal immune system include syngeneic models, where murine cancer cell lines are used, genetically engineered models and models where a known carcinogen is applied. In the presented model, no known oncogenic mutations or carcinogens are used, thus underlying the importance of inflammation in the development of cancer.

As the requirement to test novel immunotherapies and especially combination treatments is increasing, a preclinical model that takes into account tumor microenvironment and immune system would be highly useful to promote development of novel therapies to combat prostate cancer.

Aim of the Study

Aim of the present study was to reveal if there is role between the immune system and development of prostate cancer, and secondly, to validate an animal model to be utilized in immunotherapy development.

Materials and Methods

Intact 10-12 weeks old male Noble rats were s.c. implanted with slow-releasing testosterone and estradiol pellets (Innovative Research of America, Florida) for 6, 13 and 18 weeks. Estimated daily release for testosterone was 0.8 mg and for estradiol 0.08 mg. Control group animals received placebo hormone pellets. At the end of the study, prostates and selected tissues were removed and weighed. Prostate-urictra blocks were processed and serial paraffin sections were cut for histopathological evaluation. HE-staining was performed for basic histopathological analysis and immunostaining using antibodies against CD4+ and CD68 (mononuclear mouse anti-rat, Caltag Laboratories, Burlingame, Canada) was used to reveal localization of T-lymphocytes and stromal cells, respectively. CD163 clone E20 (Abcam) was stained (BiocellBio Ltd, Tampa, Florida) for M2-type macrophages.

Results

Figure 1. Hormonal treatment increased estradiol to testosterone ratio, and caused increase in weight of hormonal sensitive organs A) prostate and B) pituitary glands after all treatment periods 6, 13 and 18 weeks. C) Extent of chronic inflammation in dorsolateral prostate lobes increased concurrently with period treatment length.

Figure 2. Imbalance in hormone-miles induced inflammation in the prostate mainly in the dorsolateral site (A-D) and in the periurethral area (E-H). Inflammation was evident as perivascular (arrow in B), stromal (arrow in C) and periglandular (arrow in D) T-lymphocytes and intraluminal (arrow in D) neutrophils. In the periurethral area formation of prostatic intraepithelial neoplasia (PIN)-like lesions after 13 weeks (G) and finally adenocarcinomas (H) after 18 weeks were observed. Malignant changes were not observed in placebo-treated (E) or 6 weeks treated (F) prostates.

Figure 3. A) Inflammatory foci in prostate consisted mainly of T-lymphocytes (CD3+ cells). B) Specific subset of T-cells, cytotoxic (CD8+) cells were observed in perivascular lesions. C) Cytotoxic T-cells located also interstitially.

Figure 4. Macrophages specific for CD163 (M2 type macrophage) were present in both prostate (A) and periurethral side (B-C). A) In the prostate M2 cells were present only in the stromal compartment without inflammatory foci. M2 cells were absent in inflamed prostatic acini. M2 cells were abundant in the periurethral stromal areas but absent in B) PIN-like lesion areas or C) at the adenocarcinoma site.

Conclusions

Presence of lymphocytes in the proximity of PIN-like lesions during the early phases of prostate cancer, and their disappearance later in the adenocarcinomas, poses some interaction between stromal and adaptive immune system and cancer. The role of macrophages in the progress from prostatic inflammation towards cancerous lesions in this model remains to be revealed.

This preclinical prostate cancer model that combines immune system and cancer can be utilized when new immunotherapies, combination treatments and prevention possibilities against prostate cancer progression are developed.