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European Association of Urology

Prostate Cancer

The Mitochondrial and Autosomal Mutation Landscapes of Prostate Cancer

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Article info

Article history:
Accepted November 27, 2012
Published online ahead of print on December 5, 2012

Keywords:
Prostate cancer
Exome sequencing
Proliferation
Copy number
Indolence

Abstract

Background: Prostate cancer (PCa) is the most common cancer in men. PCa is strongly age associated; low death rates in surveillance cohorts call into question the widespread use of surgery, which leads to overtreatment and a reduction in quality of life. There is a great need to increase the understanding of tumor characteristics in the context of disease progression.
Objective: To perform the first multigenome investigation of PCa through analysis of both autosomal and mitochondrial DNA, and to integrate exome sequencing data, and RNA sequencing and copy-number alteration (CNA) data to investigate how various different tumor characteristics, commonly analyzed separately, are interconnected.
Design, setting, and participants: Exome sequencing was applied to 64 tumor samples from 55 PCa patients with varying stage and grade. Integrated analysis was performed on a core set of 50 tumors from which exome sequencing, CNA, and RNA sequencing data were available.
Outcome measurements and statistical analysis: Genes, mutated at a significantly higher rate relative to a genomic background, were identified. In addition, mitochondrial and autosomal mutation rates were correlated to CNAs and proliferation, assessed as a cell cycle gene expression signature.
Results and limitations: Genes not previously reported to be significantly mutated in PCa, such as cell division cycle 27 homolog (*Saccharomyces cerevisiae*) (*CDC27*), myeloid/lymphoid or mixed-lineage leukemia 3 (*MLL3*), lysine (K)-specific demethylase 6A (*KDM6A*), and kinesin family member 5A (*KIF5A*) were identified. The mutation rate in the mitochondrial genome was 55 times higher than that of the autosomes. Multilevel analysis demonstrated a tight correlation between high reactive-oxygen exposure, chromosomal damage, high proliferation, and in parallel, a transition from multiclonal indolent primary PCa to monoclonal aggressive disease. As we only performed targeted sequence analysis; copy-number neutral rearrangements recently described for PCa were not accounted for.
Conclusions: The mitochondrial genome displays an elevated mutation rate compared to the autosomal chromosomes. By integrated analysis, we demonstrated that different tumor characteristics are interconnected, providing an increased understanding of PCa etiology.

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0302-2838/$ – see back matter
http://dx.doi.org/10.1016/j.eururo.2012.11.053

1. Introduction

Prostate cancer (PCa) is the most common male cancer in Europe and North America. While highly prevalent, the disease progresses only in a small proportion of patients, thus the majority of patients under active surveillance will eventually die of other causes [1]. As PCa is found in most diseased, elderly men without clinical symptoms [2], it is unlikely that early high-frequency events, such as the transmembrane protease, serine 2–v-ets erythroblastosis virus E26 oncogene homolog (*TMPRSS2-ERG*) gene fusion or speckle-type POZ protein (*SPOP*) mutations, reported to be present in prostatic intraepithelial neoplasia [3,4], will be associated with aggressive disease progression. For the *TMPRSS2-ERG* gene fusion, this was the conclusion of the largest study performed to date [5], in which the gene fusion was not associated with biochemical relapse or PCa death. Still, only lung cancer has higher death rates than PCa, and consequently there is considerable interest in identifying markers of prognosis and molecular drivers of progression.

Transcript profiling of PCa has led to the development of a cell cycle gene signature reportedly predictive of poor prognosis [6]. A perturbed genome with many copy number alterations (CNAs) has also been reported to predict poor outcome in PCa [7]. Furthermore, exome sequencing has been used in PCa to identify candidate drivers [4,8]. Although, these studies bring valuable information in the pursuit of candidate biomarkers harboring information on disease outcome, it is not clear how these tumor characteristics, commonly analyzed separately, are interconnected as PCa progresses from organ-confined, indolent disease to metastatic, late-stage disease.

In this study, we used exome sequencing, copy-number profiling, and RNA sequencing in a set of 64 tumor samples from 55 patients (Table 1, Supplementary Table 1). By aligning sequencing data to both the nuclear and mitochondrial genomes, we have generated the first multigenome description of prostate tumors. By correlating multiple data types, we identified associations between changes in the mutational burden, copy-number variation, cell cycle gene signature, and clonal complexity not previously described for PCa.

2. Patients and methods

2.1. Clinical data and tissues

Information on handling of tissues and extractions can be found in the Supplementary Text. All tumor samples were harvested from men who underwent radical prostatectomy at the Karolinska University Hospital in Stockholm, except SWE-55; primary tumor and bone metastasis samples were obtained from this patient through biopsy procedures. All tumor tissues contained ≥70% tumor cells except SWE-55A, approximately 50% of which was composed of tumor cells. To retrieve somatic mutational and CNA profiles, all tumor tissues were compared to a germline DNA source retrieved from whole blood or pathologically normal prostate tissue (examined by author L.E.). Clinical data (Table 1) were collected with informed consent.

The following disease definitions were used: aggressive disease: at least one postsurgery prostate-specific antigen (PSA) value >10 ng/ml; recurrent disease: one postsurgery PSA value >0.2 ng/ml with a subsequent increase; nonrecurrent disease: fewer than two PSA values >0.2 ng/ml after surgery; unknown: this category was used for individuals with adjuvant treatment and who, therefore, did not fit the other described categories (eg, SWE-17). Despite a positive surgical margin, the PSA level decreased to <0.05 ng/ml. Following an increase in PSA during 1 yr, from <0.05 to 0.13 ng/ml, hormone and radiation treatment brought the PSA level down to <0.05.

Table 1 – Clinical characteristics

Clinical characteristics	Aggressive disease*	Recurrent disease	Nonrecurrent disease**	Unknown
Subjects, no.	8	15	25	6
Age at surgery, yr, mean (SD)	62.9 (4.9)	65.7 (4.4)	63.9 (5.7)	62.8 (4.5)
Follow-up, yr, mean (SD)	1.7 (1.8)	2.4 (1.9)	5.2 (1.8)	6.2 (1.1)
Maximum preoperative PSA value, ng/ml, mean (SD)	6.9 (2.1)	13.1 (7.2)	10.4 (6.5)	16.5 (8.2)
Maximum postoperative PSA value, ng/ml, mean (SD)	453 (1068.3)	0.9 (0.7)	0.1 (0.1)	1.1 (2.0)
Gleason score				
≤5	–	–	1	–
6	–	–	11	–
3 + 4 = 7	1	8	9	4
4 + 3 = 7	1	7	2	1
8	1	–	1	1
≥9	5	–	1	–
Missing data	–	–	–	–
Tumor stage				
T0, T1, Tx	1	–	1	–
T2	3	6	15	3
T3	4	9	9	3
T4	–	–	–	–
Missing data	–	–	–	–

SD = standard deviation; PSA = prostate-specific antigen.

* Aggressive/recurrent disease = time from surgery to the first PSA increase.

** Nonrecurrent/unknown disease = time from surgery to the most recent PSA value. Note, radical prostatectomy was not performed on patient SWE-55. This subject is therefore not included in the table. The primary tumor biopsy was taken before treatment (age 58, Gleason score 9). The primary tumor biopsy was taken before treatment (age 58, Gleason score 9). The metastasis sample was obtained 7.6 yr later after treatment with luteinizing hormone-releasing hormone agonist, Casodex, and Taxotere.