ELSEVIER

Urologic Oncology: Seminars and Original Investigations 26 (2008) 158–165

UROLOGIC ONCOLOGY

Original article

Tumor necrosis factor-related apoptosis inducing ligand-R4 decoy receptor expression is correlated with high Gleason scores, prostate-specific antigen recurrence, and decreased survival in patients with prostate carcinoma☆

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Received 17 October 2006; received in revised form 16 January 2007; accepted 18 January 2007

Abstract

Objective: Tumor necrosis factor-related apoptosis inducing ligand (TRAIL) has recently been investigated because of its ability to selectively kill cancer cells. Despite recent publications mainly focusing on TRAIL resistance in cancer cells, little is known about how TRAIL contributes to the carcinogenesis process. Because the expression patterns of TRAIL and its receptors in patients with prostate carcinoma have recently been reported, this study investigated the significance of TRAIL and TRAIL receptor expression in connection to serum prostate-specific antigen (PSA) and Gleason scoring.

Materials and methods: A total of 98 patients were included in the study. Gleason scores, PSA, TRAIL, and TRAIL receptor expressions were used for the comparison purposes. The Spearman rho correlation test was administered to reveal the correlations among the variants. The Kruskal Wallis-Mann Whitney *U* or Friedman-Wilcoxon signed ranks test determined the statistical significance between the pairs. Multinomial and/or multiple binary logistic regression analyses were deployed to test whether TRAIL markers were independent variables to predict the prognosis of prostate cancer. Kaplan-Meier and log-rank tests were used to determine the survival rates.

Results: High-serum PSA levels were correlated with higher levels of TRAIL and TRAIL receptor expressions. Patients with high Gleason scores had higher levels of TRAIL-R4 decoy receptor expression but lower levels of TRAIL death ligand expression.

Conclusions: TRAIL-R4 decoy receptor expression is strongly correlated with PSA recurrence, which is suggestive of poor prognosis. High levels of TRAIL-R4 expression but low levels of TRAIL death ligand expression are connected to decreased survival.

Keywords: Tumor necrosis factor-related apoptosis inducing ligand; Gleason scoring; Prostate-specific antigen recurrence; Prostate cancer

1. Introduction

Prostate cancer is 1 of the most frequently diagnosed malignancies among men in the Western world, and 29,900 cases of death are expected this year in the United States alone [1]. The proper assessment of the prostate cancer progression is a pivotal step when counseling the patient for the curative versus palliative therapy. Stage, Gleason score, and serum prostate-specific antigen (PSA) are all well-established prognostic factors that are routinely used in the clinical decision-making process [2]. Despite the fact that numerous studies linked high-serum PSA levels to the clinically advanced stages of prostate carcinoma [3], in most cases, serum PSA measurement alone does not provide an accurate assessment of the disease progression for a given patient [4]. For example, preoperative evaluation of the serum PSA in patients with prostate cancer is confounded by both the volume of the benign prostate tissue present [5]

☆ This study was supported by Akdeniz University Scientific Research Project Administration Division Grant.

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1078-1439/08/$ – see front matter © 2008 Elsevier Inc. All rights reserved.
doi:10.1016/j.urolonc.2007.01.022

and also the tumor grade [6]. Of the many histologic grading systems introduced to help to predict the pathologic stage and prognosis of prostate cancer, the most commonly used is the Gleason system [7,8]. Despite this fact, even Gleason score alone is insufficient to predict accurately the pathologic stage because of a previously described phenomenon of histologic upgrading from biopsy to prostatectomy specimens [8,9]. Currently, earlier staging systems of prostate carcinoma mainly rely on the digital rectal examination (DRE). However, its relative lack of sensitivity (52%) limits DRE [3]. Thus, traditional prognostic markers (grade, clinical stage, and pretreatment PSA) are of limited predictive value for the pathologic staging of the prostate carcinoma. Consequently, additional markers are strongly needed to define high-risk patients more accurately for both the pathologic staging and forthcoming therapies of prostate carcinoma.

Genes involved in the cellular proliferation such as tumor repressor genes (phosphatase and tensin homolog, etc.) [10,11] and those genes controlling the cell death [12] are very attractive for investigation because of their potential to be used as prognostic markers to predict the disease progression. One such marker is the tumor necrosis factor-related apoptosis inducing ligand (TRAIL), which is an apoptosis-inducing member of the tumor necrosis factor family [13]. Investigations concerning TRAIL have become very popular because of its therapeutic potential as a result of selective apoptosis inducing properties on cancer cells [14]. However, it is still unclear how TRAIL and TRAIL receptor expression profiles influence the carcinogenesis process. TRAIL can interact with 4 distinct receptors. Two of these receptors, DR4 (TRAIL-R1) and DR5 (TRAIL-R2), are membrane-spanning proteins containing intracellular death domains essential for the transmission of the death signal upon TRAIL binding and receptor trimerization. Two other membrane receptors, DcR1 (TRAIL-R3) and DcR2 (TRAIL-R4), can also bind TRAIL but lack death domains and are unable to induce cell death [15]. Because the presence or absence of TRAIL decoy receptors were connected to the sensitivity of cancer cells to apoptotic ligands [16–18], the modulation of TRAIL and TRAIL receptor expression might be essential for the progression of prostate cancer [19]. Therefore, the aim of this study was to investigate the potential connection of TRAIL and its receptors to the currently known prognostic factors (serum PSA and Gleason scoring) for a better assessment of prostate carcinogenesis.

2. Materials and methods

2.1. Clinical assessment of patients with prostate cancer

A total of 44 patients with benign prostate hyperplasia (BPH), 28 with organ-confined prostate carcinoma and 26 with advanced prostate carcinoma, admitted to the Urology Clinic of Akdeniz University Hospitals were included in the study. Pretreatment PSA levels were obtained from patient's serum in the Central Laboratory of Akdeniz University Hospitals. Pathologists determined the Gleason score for each patient. Patients with advanced prostate cancer possessed clinical and radiologic evidence of metastatic disease. Prostate tissues were acquired from patients undergoing radical prostatectomy for the organ-confined disease. Other tissues were attained through transurethral resection of prostate (TURP) for patients with BPH and transrectal ultrasound-guided biopsy for patients with advanced prostate carcinoma. Before the TURP operation, patients with BPH received α-blockers, while no patient with prostate cancer received neoadjuvant or adjuvant therapy, including androgen ablation therapy or chemotherapy/radiation, before surgery. Clinical and pathologic stages were assigned according to the tumor-nodes-metastasis prostate cancer staging system [20]. Disease progression for patients with advanced prostate carcinoma was defined as the appearance of new lesion(s), and/or an increase of $\geq$25% of measurable metastases, and/or the appearance of new foci on a radionuclide bone scan, and/or 3 consecutive increases in PSA concentration at least 1 week apart in the presence of testosterone castrate level ($<$50 ng/ml) of patients with metastatic disease. A postoperative total PSA level of $\geq$0.4 ng/ml and increasing was considered as evidence of biochemical (PSA) recurrence for patients with organ-confined prostate carcinoma [21].

2.2. Histologic grading of the prostate tissue and specimen processing

The processing of prostate tissue samples was performed as previously described [22]. Patients were given a designated Gleason grading score based on the specimens obtained through the radical prostatectomy, transrectal ultrasound-guided biopsy, and TURP [7]. Briefly, the Gleason grading system is based on a low-power microscopic description of the histologic architecture of cancer. A Gleason grade of 1–5 was assigned as a primary grade (pattern occupying in the largest area of the specimen) and as a secondary grade (pattern occupying the second-largest area). Adding the primary and secondary grades determined a Gleason score (2–10). In this study, patients were separated into 2 groups based on Gleason scoring, as those with Gleason score $\geq$7 and those with Gleason scores $<$7 [23].

2.3. Immunohistochemical scoring of TRAIL and TRAIL receptors, and the statistical analysis

The immune staining procedures of prostate sections and the scores were described elsewhere [18,19]. Two independent pathologists who were blinded to the names of the antibodies used for the staining performed immunohistochemical scoring. To explain briefly, both the intensity and the marker distribution (percentage of positively stained