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Urologic Oncology: Seminars and Original Investigations ▮ (2017) ▮▮▮–▮▮▮

UROLOGIC ONCOLOGY

Review article

# Insight into novel biomarkers in penile cancer: Redefining the present and future treatment paradigm?

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Received 10 March 2017; received in revised form 20 July 2017; accepted 10 October 2017

**Abstract**

**Introduction:** Biomarkers are increasingly used in the diagnosis and management of various malignancies. Selected biomarkers may also play a role in management of certain cases of penile carcinoma. In this article, we provide an overview of the clinical role of such markers in the management of penile cancer.

**Method:** This is a nonsystematic review of relevant literature assessing biomarkers in penile carcinoma.

**Results:** Evidence of infections with human papillomavirus and its surrogate markers may have important prognostic value in patients with localized or metastatic penile cancer. Squamous cell carcinoma antigen, p53, C-reactive protein, Ki-67, proliferating cell nuclear antigen, cyclin D1, as well as other markers have been studied with various degree of evidence in support of clinical utility in penile cancer.

**Conclusions:** No single marker may have all the answers, and future research should focus on genomic analysis of individual penile tumors, attempting to identify specific targets for treatment. 

*Keywords:* Penile cancer; Biomarkers; p16 ink4a; HPV

## 1. Introduction

In 1998, the National Institutes of Health Biomarkers Definitions Working Group defined a biomarker as "a characteristic that is objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes, or pharmacologic responses to a therapeutic intervention" [1]. Based on this definition, there have been many such biomarkers which have been identified across many different disease states.

Among genitourinary malignancies, penile cancer is unique in that a specific viral infection has been linked to the disease process, providing additional potential biomarkers. Biomarkers in penile cancer have the potential to serve as an important prognostic tool to predict disease progression, recurrence, and development of metastasis. Data, however, are still not rigorous enough to include the routine use of biomarkers in the diagnosis and management of penile malignancy.

In the present work, we review this timely topic highlighting some of our work and that of others in this field. A summary of several biomarkers pertinent to this topic are highlighted in Table 1.

## 2. Methods

This is not a systematic review of the topic. We performed Medline databases searches with keywords "penile carcinoma" and "biomarkers," and reviewed the results with inclusion of any relevant articles. Emphasis was placed on the most recent review articles. In addition, the reference list of these publications were also searched for other relevant literature.

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http://dx.doi.org/10.1016/j.urolonc.2017.10.010
1078-1439/

Table
Biomarkers in penile cancer

| Biomarker | Number of studies | Function | Prognosis |
|---|---|---|---|
| SCCAg | 5 | Tumor-associated glycoprotein | Elevated levels predictive of disease burden including nodal and metastatic disease; can be used to monitor disease progression and response to treatment |
| p53 | 6 | Tumor suppressor gene | Expression indicated higher risk of LN metastasis, disease progression, and worse DSS |
| CRP | 3 | Proinflammatory marker | Elevated plasma levels found more often in patients with advanced tumor stage, positive nodal disease, and worse DSS |
| Ki-67 | 4 | Marker for tumor cell proliferation in the cell cycle | Labeling correlated with higher tumor grade, advanced local tumor stage, a greater risk of nodal metastasis, and clinical disease progression |
| PCNA | 2 | Marker of cell proliferation essential for replication | Expression was associated with presence of nodal metastasis |
| Cyclin D1 | 2 | Regulates progression of cells through G1-phase of the cell cycle | No clear prognostic value; implicated in tumor differentiation |
| p16ink4a | 5 | Surrogate marker for high-risk HPV infection | Positivity was associated with less tumor invasion, lower risk of disease recurrence, and possibly better survival |
| E-cadherin | 1 | Maintains cellular adhesion and signal transduction | Immunoreactivity was associated with a greater risk of LN metastasis |
| MMP-2 and MMP-9 | 1 | Degrades the basement membrane of a cell | Immunoreactivity was associated with a greater risk of disease recurrence |
| Fox-P3 | 1 | Oversees the development and function of regulatory T cells | Increased levels correlated to a lower inflammatory infiltrate worse OS |
| ARID1A | 1 | Involved in chromatin remodeling | Higher expression was associated with a higher histologic grade |

DSS = disease-specific survival.

## 3. Human papillomavirus and surrogate markers

Improved understanding of the etiological factors contributing to the development of penile cancer has allowed for the utilization of newer biomarkers. Totally, 23% to 50% of cases of penile carcinoma are seen in presence of human papillomavirus (HPV) infections, and this is likely unrelated to the patients' age [2–7]. In penile cancer, HPV infection causes a disruption in the normal regulatory cell pathways including apoptosis through a process mediated by HPV E7 and E6 oncoproteins, which bind to the host retinoblastoma and p53 proteins. The subsequent inactivation of these cell-cycle check points leads to abnormal cellular proliferation. The hallmark of HPV-induced penile cancer is the accumulation of p16ink4a in affected cells. Immunohistochemical staining can detect the overexpression of p16ink4a, and this can serve as a marker for transcriptionally active HPV infection. P16ink4a overexpression is only seen in high-risk HPV genotypes [8,9]. Penile cancers where HPV infection is not present are due to p53 mutations with penile intraepithelial neoplasia being proposed as a precursor lesion. Penile intraepithelial neoplasia often demonstrates p53 overexpression in absence of p16ink4a expression [8].

We have recently have demonstrated a relationship between HPV infections and clinical outcomes [10]. In a cohort of 57 patients and using immunohistochemical staining for p16ink4a and in situ hybridization (ISH) for high-risk HPV, we identified that 40% and 42% of patients were p16ink4a and HPV positive, respectively, with only relatively minor discordance between HPV-ISH and p16ink4a immunohistochemistry results. This supported the use of p16ink4a expression as a surrogate marker for HPV infection. Additionally, only 1 patient was both p53 and p16ink4a positive, supporting the use of p53 expression as a surrogate marker for the absence of HPV infection. In our study cohort, the majority (65%) of patients who were p53 positive had positive lymph node (LN) disease at the time of surgery. In fact, in patients who were p16ink4a negative, p53 positivity was an independent predictor of nodal metastases (odds ratio = 4.4, 95% CI: 1.04–18.6). On Kaplan-Meier survival analysis, the unadjusted estimated overall survival (OS) was not significantly longer in p16ink4a positive vs. negative patients (median OS: 75 vs. 27 months, respectively, $P = 0.27$), and median cancer specific survival (CSS) was not reached ($P = 0.16$, Fig. 1). We also demonstrated that in p53 negative men, p16ink4a positivity had a potentially positive influence on CSS ($P = 0.07$). In pathologically node-positive patients, CSS was significantly worse in the few patients with both negative p16ink4a and p53 expression (8 vs. 34 months, respectively, $P = 0.01$, Fig. 2). More significantly, on multivariable Cox proportional hazard model and after adjusting for adjuvant chemotherapy and nodal status, we demonstrated that p16ink4a positivity was a significant predictor of improved CSS compared to lack of p16ink4a expression (hazard ratio [HR] = 0.36, 95% CI: 0.13–0.99).

Tang et al. [5] reported on 119 penile carcinomas who had immunohistochemistry staining for p16ink4a. Approximately, 50% of cases were p16ink4a positive. They did not