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Molecular and genetic prognostic factors of prostate cancer

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Abstract Prostate cancer is the most commonly diagnosed cancer in Western males, responsible for 3% of all deaths in men over 55 years of age and second only to lung cancer as a cause of cancer death. Biomarkers have become an important diagnostic tool in prostate cancer. The discovery of the serum marker prostate-specific antigen (PSA) significantly facilitated the detection and management of prostate cancer. As we enter into the post-genomics era, novel biomarkers of prostate cancer of therapeutic significance will invariably emerge. Here we review a series of existing and emerging molecular-based prognostic markers particularly with radiotherapy.

Keywords Prostate cancer · Radiotherapy · Prognosis · Molecular/genetic biomarkers · Cell cycle · Apoptosis · Angiogenesis · Signal transduction · Invasion/adhesion · Tumor suppressor genes

Morphology-based approaches, especially Gleason scoring, have provided clinicians with important prognostic information, especially when combined with clinical parameters of prostate-specific antigen (PSA) and T stage [1, 2, 3, 4, 5, 6, 7]. However, the prognostic value of the Gleason score is limited by the fact that the vast majority of prostate cancer patients present with moderately differentiated tumors (e.g., Gleason score of 6) in the PSA era, limiting the prognostic utility of morphologic features.

Recent successes in functional genomics and proteomics have served to cultivate the growing interest in discovering more molecular-based prognostic factors that could be utilized to assay the original needle biopsy specimen to tailor the primary treatment for individual prostate cancer patients [8, 9, 10, 12]. As targeted therapy in oncology becomes increasingly powerful, there is a significant interest in finding prognostic markers in prostate cancer that could be used as targets for novel biotherapies. Many molecular- and genetic-based biomarkers have been discovered over the last 2 decades and are summarized in Table 1 and also in Abate-Shen and Shen [89]. This article discusses their prognostic significance in prostate cancer treatment.

Cell cycle makers

Cell cycle regulators have been found to associate with adverse outcome in prostate cancer [11]. For instance, cyclin D1 appears to have an impact on both breast and prostate tumorigenesis by mediating hormone receptor signaling [11]. Cyclin D1 overexpression is significantly associated with the subsequent development of distant metastasis in prostate cancers [24]. The Cip/Kip and INK4 groups of cyclin-dependent kinase inhibitors that regulate the G1 to S phase transition of the cell cycle [11] have been found to play roles in prostate cancer [11, 25, 26, 27, 28, 29]. p21 immunoreactivity was found to be associated with improved survival [25, 26, 27]. Loss of p27 expression has been linked with adverse disease outcome in a number of studies [11, 28, 29]. In prostate carcinoma, the tumor suppressor gene CDKN2/MTS1 (p16 INK4) has a very low frequency of point mutations, but deletions of 9p21 and inactivation by promoter methylation are observed more frequently. In a recent study, Halvorsen et al. evaluated the expression of p16 and CDK4 proteins and their prognostic significance in patients with clinically localized prostate carcinoma treated surgically [30]. In their univariate survival analysis of the first 5 years, high levels of p16 protein

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Table 1 A list of prognostic markers categorized based on their biological functions and discussed in this review. Category I: markers that have been proven to be reliable in prognosis and generally used in patient management. Category II: markers that show promise as a result of extensively biological and/or clinical

studies but with few clinical outcome studies. Category III: markers that have some scientific evidence to support their adoption as diagnostic or prognostic agents but are not currently recommended; or those factors with uncertain significance

Type of marker	Prognostic factor	CAP category	Reference
Cell cycle	cyclin D1	III	[11, 24]
	p21	III	[25, 26, 27]
	p27	III	[11, 28, 29]
	p34cdc2	III	[31]
	Ki-67 (MIB-1)	III	[16, 17, 18, 19, 20, 21]
Angiogenesis	Tumor vascularity	III	[42]
	Microvessel density	III	[43, 44]
	VEGF	III	[49, 50]
	bFGF	III	[49, 50]
	Endothelin-1	III	[119]
Apoptosis	Bcl-2	III	[71]
	Bax	III	[71]
	Bcl-x	III	[71]
Oncogenes + tumor suppressor	<i>ras. myc</i>	III	[11, 87]
	p16(INK4)	III	[30]
	p53	III	[90]
	PTEN/MMAC1	III	[101, 102]
Tumor invasion + cell adhesion	Cathepsin D	III	[82, 83]
	Metalloproteases	III	[85]
	E-cadherin	III	[74, 75, 76, 77, 78, 79, 80, 81]
Signal transduction	Caveolin-1	III	[107, 108, 109, 111]
	HER-2/ <i>neu</i>	III	[11, 32, 92]
	EGF	III	[72]
	EGFR	III	[116]
	TGF- β	III	[11, 72, 73]
	MAPK&MKK4	III	[14, 105]
Epigenetics	DNA ploidy	II	[32, 33, 34, 35, 36, 37, 38, 39]
Other types	GST- τ	III	[91]
	Syndecan-1	III	[112, 113, 114, 115, 117, 122]
	EZH2	III	[110]

expression, tumor greatest dimension, WHO histologic grade, capsular penetration, seminal vesicle invasion, positive surgical margins, lymph node involvement, and preoperative serum prostate-specific antigen > 20 ng/ml all were significant predictors of biochemical failure. In their multivariate survival analysis, high p16 protein expression, age, WHO histologic grade, capsular penetration, and seminal vesicle involvement remained as independent predictors of biochemical failure. This study suggests that elevated expression of p16 protein, but not CDK4 protein, may play a significant role in the development of prostate carcinoma and represent an independent predictor of biochemical failure after radical prostatectomy. However, loss of p16 expression was found to be associated with adverse outcome (e.g., reduced overall and disease-specific survival) in patients with locally advanced prostate cancer treated by radiation therapy \pm neoadjuvant hormones [116]. The observed discrepancy in the prognostic significance of p16 expression between radiation and surgery-treated patients suggests that the type of treatment delivered may be an important factor determining the significance of a given molecular marker.

Overexpression of p34^{cdc2} cyclin-dependent kinase in the S to G₂M transition of the cell cycle was shown to

have connection with aggressive high-grade disease with increased incidence of biochemical failure after primary therapy [31]. As for the cell proliferation markers, immunohistochemical analysis using MIB-1/Ki-67 antibody is very useful for measuring cell cycle progression in human tissues [16, 17]. It is usually accepted that >16–20% MIB-1 staining is associated with a high proliferation rate and an adverse prognosis although clinically defined proliferation ranges remain under study for prostate cancer [16, 17, 18, 19, 20, 21]. Primary therapy failure has been attributed to MIB-1 overexpression, which has appeared useful in predicting prognosis even in patients with existing lymph node involvement [15, 22].

To establish the significance of MIB-1/Ki-67 staining as a prognostic marker of treatment outcome for prostate cancer patients treated by radiotherapy, Pollack et al. carried out a study with pretreatment archival prostate biopsy tumor tissue available from stage T1-T4 prostate cancer patients treated with external beam radiotherapy [23]. The percentages of Ki-67-positive tumor cells, the Ki-67 labeling index (Ki-67 LI), were determined using immunohistochemical staining for MIB1. Experimental analyses indicated that, for all patients, mean Ki-67 LI levels were higher with stage

T3/T4 disease, Gleason 7–10 disease, and in those that developed treatment failure. Comparable relationships were also observed when the Ki-67 LI was dichotomized into low and high groups. Their analysis using the approach of Cox proportional hazards regression demonstrated that dichotomized Ki-67 LI was an independent correlate of bNED (biochemical no evidence of disease) survival, along with pretreatment PSA, Gleason score, and clinical stage. This concludes that (1) Ki-67 LI could be used as a reliable independent predictor of failure after radiotherapy using biochemical criteria and (2) such a prognostic marker appears equally valuable for patients diagnosed by transurethral resection of the prostate (TURP) or needle biopsy

Tumor suppressor genes

Many studies have demonstrated that p53 or genes that affect p53 function are mutated in most, if not all, human tumors. Several studies, however, have shown that p53 mutations are sporadic in prostate cancer and are associated with advanced disease [88, 92, 93, 99]. Recently, Grignon et al. [90] evaluated the prognostic value of p53 protein expression at unusual levels in the tumors of patients with locally advanced prostate cancer under either external beam radiation therapy alone or total androgen blockade before and during the radiation therapy. The study was undertaken through a population consisting of a subset of patients entered in Radiation Therapy Oncology Group protocol 8610 via immunohistochemical detection of abnormal p53 protein in pretreatment specimens. Relationships between p53 protein expression level and the time to local progression, the incidence of distant metastases, progression-free survival, and overall survival were assessed. Abnormal p53 protein expression, which is often an indicator of mutated p53, was observed in the tumors of 18% of patients. A statistically significant connection was revealed between abnormal p53 expression and elevated incidence of distant metastases, reduced progression-free survival, and reduced overall survival; however, no association was found between abnormal p53 protein expression and the time to local progression, which were independent of the Gleason score and clinical stage. For patients under both radiation therapy and hormone therapy, those with tumors displaying abnormal p53 protein expression underwent a reduced time to the development of distant metastases; for patients on radiation therapy alone, the time to distant metastases was independent of p53 protein expression status. They concluded that p53 protein expression level could yield significant, independent prognostic information concerning the development of distant metastases, progression-free survival, and overall survival for patients with locally advanced prostate cancer under ongoing radiation therapy. Therefore, the relationships of radiation and hormone therapies and aberrant p53 protein expression may well shed light on experimental evidence

that radiation therapy and/or hormone therapy work, to some degrees, by the p53-dependent apoptosis.

The subsequent biological changes in recurrent prostate carcinomas following radiation treatment are far from being completely appreciated. In a different study, Cheng and his colleagues [91] demonstrated that p53 protein overexpression is associated with increased cell proliferation in patients with locally recurrent prostate carcinoma after radiation therapy. Their study determined the level of p53 protein overexpression and its association with cellular proliferation (Ki-67 LI), glutathione S-transferase- π (GST- π) expression, and other clinical pathologic findings in patients with locally persistent prostate carcinoma after radiation therapy. The respective expression status of p53 nuclear accumulation, cellular proliferation activity, and GST- π was investigated in patients with persistent or recurrent prostate carcinoma post radiation therapy. Experimental analysis revealed that p53 protein overexpression was associated with increased cell proliferation. A substantial proportion of recurrent cancer also showed GST- π immunoreactivity. However, no apparent correlation was seen between p53 protein overexpression, cellular proliferation (Ki-67 LI), or GST- π expression and Gleason score, pathologic stage, DNA ploidy, or patient outcome. Analysis also established an inverse correlation between GST- π expression and Gleason score. The majority of prostate carcinomas (95%) were proliferative whereas concurrent prostatic intraepithelial neoplasia (PIN) had a lower Ki-67 labeling index. Sixty eight percent of the concurrent PIN demonstrated p53 immunoreactivity. A trend toward adverse clinical outcome was observed in patients with a higher Ki-67 LI in recurrent cancer. In their study, recurrent cancers were biologically aggressive following radiation therapy, which may represent selective persistence and regrowth of prognostically unfavorable tumor clonogens or step-wise clonogenic progression.

PTEN/MMAC1

The PTEN/MMAC1 phosphatase is a tumor suppressor gene implicated in a wide range of human cancers [95, 96, 97]. The PTEN/MMAC1 tumor suppressor gene is deleted or mutated in a wide variety of cancers including prostate cancer cell lines, xenografts, and clinical samples [11, 98, 99]. Sawyers and his colleagues provided biochemical and functional evidence that PTEN/MMAC1 acted as a negative regulator of the phosphoinositide 3-kinase (PI3-kinase)/Akt pathway [100]. PTEN/MMAC1 was shown to exhibit the capacity of activating endogenous Akt in cells and inhibiting phosphorylation of 4E-BP1, a downstream target of the PI3-kinase/Akt pathway involved in protein translation, whereas a catalytically inactive, dominant negative PTEN/MMAC1 mutant enhances 4E-BP1 phosphorylation. Moreover, PTEN/MMAC1 suppresses gene expression in a way that was salvaged by Akt but not

PI3-kinase. More significantly, elevated levels of Akt activation were detected in human prostate cancer cell lines and xenografts lacking PTEN/MMAC1 expression when compared with PTEN/MMAC1-positive prostate tumors or normal prostate tissue. PTEN expression loss has been associated with downregulation of the cyclin-dependent kinase inhibitor p27 and adverse outcome [101] and increasing grade and stage [102] in prostate cancer.

Caveolin-1

Caveolin-1 protein was initially discovered by differential gene expression display technique and has been associated with both signal transduction and cell motility in prostate cancer [41, 107, 108]. Recently, Yang et al. [111] established that Cav-1 expression is significantly increased in primary and metastatic human prostate tumors after androgen ablation therapy. And also, Cav-1 is secreted by androgen-insensitive prostate carcinoma cells, and this secretion is regulated by steroid hormones. In cases of clinically determined, localized prostate carcinoma, they found that Cav-1 expression is a novel prognostic marker with independent predictive value of biochemical recurrence. It was observed that the 5' promoter CpG islands of Cav-1 were more methylated in tumor than in adjacent normal prostate cells implying that Cav-1 may serve as a tumor suppressor gene in prostate cancer [109].

Invasion and cell adhesion markers

Tumor invasion associated proteases

Cathepsin D is a lysosomal protease and autocrine mitogen and has been demonstrated to associate with prognosis in breast cancer [82]. In prostate cancer increased tumor cathepsin D immunoreactivity has been connected with pathologic stage [83], tumor grade, and DNA content [84]. Elevated serum levels of soluble urokinase plasminogen activator receptor have been connected with progressive prostate cancer. Localization of matrix metalloproteases has been linked to prostate cancer development [85]. Upregulated expression of tumor collagenases has also been linked to adverse disease outcome [13].

Cell adhesion molecules

E-cadherin is a cell adhesion molecule whose aberrant cellular expression has been linked to adverse disease outcome in prostate cancer [74, 75, 76, 77, 78, 79, 80, 81]. For instance, reduced expression of E-cadherin has been correlated to high tumor grade and aneuploidy [74]. A study by Sandberg a decade ago implied that a major chromosomal deletion on chromosome 16 may be a

major event in the development of prostate cancer [79]. Further investigation showed that this deletion event may involve the E-cadherin gene; its expression product E-cadherin protein may serve as a tumor suppressor protein. As a result of the establishment of the relationship of β -catenin degradation and association with the APC (adenomatosis polyposis coli) gene, the E-cadherin interaction with β -catenin has been a subject of great interest [11, 81].

Signal transduction markers

Dominant oncogenes

The contributions of dominant oncogenes in prostate cancer development and progression appear to be diminutive in view of their significance in adenocarcinomas of the respiratory and gastrointestinal tract [40, 86]. For instance, the *ras* genes, frequently mutated in the malignancies of the gastrointestinal, hepatobiliary, and respiratory tracts, often remain unaltered in human prostate cancers or cell lines or experimental models [87]. Although *Myc* gene mutations may be associated with development of hormone refractory disease [11], study has also shown that the amplification of the *myc* gene was implicated in prostate cancer but without any direct linkage to disease progression [87].

HER-2 oncoprotein overexpression

For breast cancer patients, HER-2 oncoprotein overexpression has been linked to adverse outcome in breast cancer [92]. This fact has led to the clinical testing of HER-2 as a standard of practice status for guiding treatment with various therapies. For prostate cancer, previous studies found that overexpression of HER-2 was associated with an adverse outcome [11, 32, 92]. HER-2 mRNA levels have recently been correlated with metastatic disease and androgen-independent hormone refractory progressive disease [93]. More significantly, in a recent study, the prognostic relevance of Her-2 protein expression in patients undergoing curative radiotherapy (RT) was compared to the traditional prognostic factors such as pretreatment PSA levels, biopsy Gleason score, and T category of the primary tumor [94]. The prognostic relevance of Her-2 expression was univariately associated with adverse outcome in terms of biochemical or clinical progression-free survival (B/C-PFS), clinical progression-free survival (C-PFS) and disease-specific survival (DSS). Her-2 expression, T category, and Gleason score were independently associated with C-PFS, whereas only Her-2 expression and Gleason score were associated with DSS. Her-2 expression and Gleason score together discriminated two groups of patients with 5-year DSS of 95% and 79%, respectively. Pretreatment PSA levels

were associated only with B/C-PFS but not with C-PFS or DSS. This investigation demonstrates that expression of Her-2 is of prognostic relevance in localized prostate cancer undergoing RT and implicates that analysis for Her-2 might improve prognostic algorithms for clinically relevant endpoints other than biochemical relapse.

Growth factors

A recent RTOG protocol examined epidermal growth factor receptor (EGFR) expression patterns in locally advanced prostate cancer and found less than 5% expression (Chakravarti, personal communications). It has been shown that increased expression of basic fibroblast growth factor (bFGF) is linked to adverse outcome [13] and that overexpression of transforming growth factor (TGF)- β is involved in the growth of prostate cancer cell lines [11] and a reduction in disease-free survival in clinical trials [72]. In a recent interesting study [73], the transcription factor early growth response (EGR)-1 expression was monitored to determine whether it, in the primary tumor, correlates with radiation response in terms of complete local tumor control with no evidence of disease or recurrence and no evidence of metastasis; whether it, in the post-irradiated biopsies, correlates with residual tumor; and whether it correlates with the expression of EGR-1 target genes such as TP53, pRB, and Bax. The authors analyzed pretreated surgically resected paraffin-embedded primary adenocarcinomas of the prostate for the presence of EGR-1 expression and mutation, and correlated this with clinical endpoints such as serum PSA levels and current clinical status. They also analyzed post-irradiated biopsies of prostate for the presence of EGR-1 expression, and correlated these findings to the residual tumor status. They finally analyzed prospective prostate tumor specimens for EGR-1 expression and its target genes. EGR-1 expression was measured by immunohistochemistry and mutations were screened in two regions of the EGR-1 gene: trinucleotide AGC repeats in transactivation domain (TD) and poly A tract in 3'UTR. EGR-1 overexpression correlated with treatment failure. No correlation with EGR-1 overexpression and its target genes was found, which may imply that overexpressed EGR-1 may lack transactivation function. This investigation indicated that EGR-1 overexpression in the mutant form might offer an indication of clinical failure, either local recurrence or metastasis.

MAPK and MKK4

The mitogen-activated protein kinase (MAPK) cascade of protein phosphorylation and dephosphorylation has been linked to cell proliferation in prostate cancer [14]. Using an antibody specific for dually phosphorylated extracellular-regulated kinases 1 and 2, Weber's group has examined primary and metastatic prostate tumor

specimens for the presence of activated MAPK [105]. Nonneoplastic prostate tissue produced little or no staining with activated MAPK antiserum. In prostate tumors, elevated levels of activated MAPK have been associated with increasing Gleason scores and tumor stage in prostate cancer [105]. In a separate analysis, tumor samples from two patients showed no activation of MAPK before androgen ablation therapy. Following androgen ablation treatment, high levels of activated MAPK have also been detected in patients with recurrent disease [105]. This study suggested an increase in the activation of the MAPK signal transduction pathway as prostate cancer progressed to a more advanced and androgen-independent disease. Mitogen-activated protein kinase kinase 4 (MKK4) is a potential prostate cancer metastasis suppressor [106]. Downregulated MKK4 expression has been associated with increasing Gleason grade implying that MKK4 protein may be downregulated during prostate cancer progression [106].

Apoptosis markers

Bcl-2, bax, and bcl-x

The bcl-2 family, consisting of two members, bcl-2 and bax, plays a critical role in the control of apoptosis via dimerizations of bcl-2 and bax, governing the cellular apoptosis in response to genotoxic stress rather than cell cycle arrest and repair [50, 51, 52, 54, 59, 64]. Recent studies in bcl-2 family proteins have related the overexpression of the bcl-2 anti-apoptosis protein to decreased expression of the proapoptotic protein, bax and adverse outcome in prostate cancer associated with resistance to cytotoxic chemotherapy in patients with hormone-refractory disease [53, 103, 104]. It had been established a long time ago that radiation induces apoptosis [65], and overexpression of bcl-2 has been associated with resistance to radiation [66]. It has been observed recently that locally recurrent tumors can upregulate bcl-2 expression upon radiation [68, 69, 70]. Given the experimental link of bcl-2 overexpression to clinical treatment failure, it is argued that bcl-2 can be viewed as a marker of aggressive tumor attributes and as an etiologic factor of resistance to androgen ablation and radiation [48].

More relevant to radiation therapy treatment, bcl-2 overexpression is related to patient outcome after radiotherapy in the form of radiation resistance, although the results are contradictory. It was reported that patients with bcl-2-positive diagnostic biopsy results had a significantly longer DSS [55], whereas in other studies that bcl-2 overexpression was correlated with treatment failure after radiotherapy [56, 57, 58]. These existing data support further studies of pretreatment bcl-2 levels as a marker for patient stratification. Bax is a proapoptotic protein, and it was expected that its overexpression in tumors treated by

radiation would give rise to increased cell death and improved outcome compared with tumors that had normal bax expression. For bax expression, prior studies on the usefulness of bax as a pretreatment predictor of outcome after radiotherapy in patients with prostate carcinoma are limited and the results were inconsistent [56, 58]. In the first study, the expression levels of bax and bcl-2 were related inversely to freedom from recurrence and that a high bcl-2:bax ratio was correlated with poor therapeutic responsiveness [56]. Pollack and his colleagues have demonstrated that bax expression appears to be a significant correlate of bNED, independent of the conventional pretreatment factors of clinical stage, Gleason score, and PSA level [48]. Their results also indicate that altered bax expression is a strong predictor of bNED after radiotherapy and bax was a much stronger predictor of bNED and was independent of bcl-2. Both bcl-x and bax belong to the bcl-2 family of apoptosis-regulatory proteins. While bax is proapoptotic, bcl-x has both proapoptotic (bcl-xS) and antiapoptotic (bcl-xL) effects [60, 61, 62, 63, 67]. There is still no evidence that the quantification of bcl-x has predictive value, which may be related to the lack of specificity of the antibody used in the Pollack et al. study where both proapoptotic (bcl-xS) and antiapoptotic (bcl-xL) components were measured [48]. This is a significant study in the evaluation of biomarkers for prognosis of patients who are treated with radiation therapy although further investigation is needed to determine the optimal methods for staining classification and to establish the reproducibility in a larger group of patients who are treated with radiotherapy for prostate carcinoma.

Angiogenesis markers

Tumor vascularity and microvessel density

As with other solid tumors, angiogenesis is a critical component of prostate cancer development and progression. Weidner et al. [42] showed a close correlation between increased microvascular density and Gleason score in advanced prostate cancer. Tumor angiogenesis has been associated with adverse outcome in prostate cancer as assessed by microvessel counting with the observation that significantly higher microvessel counts have been found in areas of adenocarcinoma than in the benign tissues of radical prostatectomy specimens [43, 44]. The infrequency of necrosis in prostate cancer may be explained by the fact that prostate cancers encompass the greatest concentration of microvessels in the tumoral areas. While augmented, microvasculature has been correlated with the pathologic stage of the disease [43, 45, 46, 47]; microvessel density has appeared to have association with the presence of metastasis [43]. The application of microvessel counts to needle biopsies of prostate cancer—where it could be used prospectively to plan therapy—is worth further investigation.

Angiogenic growth factors

Latest analyses have focused on the use of specific angiogenic factors—such as vascular endothelial growth factor (VEGF), basic fibroblast growth factor (bFGF) and endothelin—as prognostic serum markers for prostate cancer. While VEGF is a peptide growth factor specific for the tyrosine kinase receptors, VEGF receptor-1 and -2 have been associated with growth of endothelial cells in prostate cancer [49, 50]. The levels of VEGF and bFGF in bodily fluids could serve as the indicators of the degree of tumor-related angiogenesis that occurs in a cancer patient. A reliable immunoassay for bFGF was not available until 1991 [118], and commercial assays are now available with good sensitivity for detecting bFGF in urine and blood.

Endothelin-1 (ET1) found in high concentrations in seminal fluid appeared to be another angiogenic factor important in prostate cancer development and progression [119]. It has been demonstrated that it stimulates prostate cancer cell proliferation *in vitro*, and boosts the mitogenic effects of IGF1 and other growth factors, and shows increased expression in prostate cancer primary tumor specimens and metastases [120]. Endothelins seem to play a major role in the growth and angiogenesis of prostate cancer, and in the pathophysiology of its metastasis to bone [121].

Genetic/epigenetic makers

DNA ploidy determination

Aneuploid DNA content in prostate cancer may independently predict poor prognosis for the disease [32, 33, 34, 35, 36, 37]. DNA ploidy measurements have been performed on needle biopsy specimens, and the ploidy status determined on needle biopsies has successfully correlated with the ploidy status of corresponding radical prostatectomy specimens and independently predicted disease outcome [38]. Despite such a solid establishment of these data, only a few studies have used pretreatment diagnostic material and have involved RT-treated patients. A significant analysis of RTOG 8610 in prostate cancer DNA ploidy and response to salvage hormone therapy after radiotherapy was carried out to supplement the knowledge [39].

In their retrospective study, the predictive value of DNA ploidy was assessed in patients treated under RT alone comparing with those under RT plus short-course neoadjuvant and concurrent total androgen blockade. Their results indicated that DNA ploidy was not associated with any of the other prognostic factors. It was shown that no relationship was observed between DNA ploidy and distant metastasis, and aneuploidy was independently associated with reduced overall survival. The patients treated with RT plus total androgen blockade (TAB) with nondiploid tumors had reduced survival after salvage androgen ablation.

This RTOG study clearly showed the close association of aneuploidy with shorter survival that is related to reduced response to salvage hormone therapy for those previously exposed to short-term TAB.

Repressor of gene transcription EZH2

It has been associated through gene expression profiling that the polycomb group protein enhancer of zeste homolog 2 (EZH2) was overexpressed in hormone-refractory, metastatic prostate cancer, and clinically localized prostate cancers that express higher concentrations of EZH2 showed a poorer prognosis [110]. This suggests that dysregulated expression of EZH2 may play a role in the progression of prostate cancer, which may be considered as a marker that distinguishes indolent prostate cancer from those aggressive forms [110].

Syndecan-1 identified via tissue microarray

Syndecan-1 (CD-138), a proteoglycan that has been found to be of prognostic importance in various human tumors [112, 113, 114, 115, 117] has not been analyzed in prostate cancer until very recently. Zellweger et al. tested several molecular markers on a prostate tissue microarray (TMA) [122]. The respective expression status of Ki-67, bcl-2, p53, CD-10 (neutral endopeptidase), and syndecan-1 (CD-138) was investigated by immunohistochemistry. This study showed that Gleason grade and Ki-67 labeling index (Ki-67 LI) were independent predictors of early recurrence and poor survival while Bcl-2 predicts early recurrence and p53 was associated with poor survival. Syndecan-1 overexpression also predicted early recurrence and was much associated with tumor-specific survival, high Gleason grade, Ki-67 LI, and bcl-2 overexpression. The results of this TMA study confirmed a central prognostic importance of Gleason grading and Ki-67 LI in prostate cancer, as compared to a less pronounced role of Bcl-2, and p53. More remarkable, they identified syndecan-1 as a new prognostic factor and provided evidence for an androgen-dependent regulation of CD-10.

Conclusions

Many prognostic markers have been discovered over the last 2 decades and are described in this review as summarized in Table 1. While the morphology-based factors of Gleason grade and clinical features of PSA and T stage are widely utilized as the standard of practice, the Ki-67 cell proliferation index, p53, and bcl-2 immunostaining and DNA ploidy analysis are now exploited in many laboratories as adjuncts for predicting outcome in the disease. At the same time, still many of the remaining prognostic factors listed in this review are now under evaluation to further investigate their abilities to determine prognosis and guide

the selection of therapy for prostate malignancies. Additional prognostic and predictive tests that can predict efficacy of a specific therapy will invariably become part of the standard of care. Genomic and proteomic high-throughput array strategies will help accelerate the discovery of more molecular-based diagnostic tests that potentially drive the selection of the type, intensity, and duration of treatment. This trend will help the realization of personalized medicine to the care of newly diagnosed prostate cancer patients.

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