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Human Cancer Biology

Clinical Cancer Research

Chromosome *8p* Deletions and *8q* Gains are Associated with Tumor Progression and Poor Prognosis in Prostate Cancer

Alexander T. El Gammal[1], Michael Brüchmann[1], Jozef Zustin[1], Hendrik Isbarn[2], Olaf J.C. Hellwinkel[1], Jens Köllermann[1], Guido Sauter[1], Ronald Simon[1], Waldemar Wilczak[1], Jörg Schwarz[3], Carsten Bokemeyer[4], Tim H. Brümmendorf[4], Jakob R. Izbicki[5], Emre Yekebas[5], Margit Fisch[6], Hartwig Huland[2], Markus Graefen[2], and Thorsten Schlomm[2]

Abstract

Purpose: Deletions of *8p* and gains of *8q* belong to the most frequent cytogenetic alterations in prostate cancer. The target genes of these alterations and their biological significance are unknown.

Experimental Design: To determine the relationship between chromosome 8 changes, and prostate cancer phenotype and prognosis, a set of 1.954 fully annotated prostate cancers were analyzed in a tissue microarray format by fluorescence *in situ* hybridization.

Results: Both *8p* deletions and *8q* gains increased in number during different stages of prostate cancer progression. *8p* deletions/*8q* gains were found in 26.1%/4.8% of 1,239 pT_2 cancers, 38.5%/9.8% of 379 pT_{3a} cancers, 43.5%/8.9% of 237 pT_{3b} cancers, 40.7%/14.8% of 27 pT_4 cancers, 39.1%/34.8% of 23 nodal metastases, 51.9%/33.3% of 27 bone metastases, and 45.5%/59.9% of 22 hormone refractory cancers ($P < 0.0001$ each). Both *8p* deletions and *8q* gains were also significantly associated with high Gleason grade and with each other ($P < 0.0001$ each). In primary tumors, *8p* deletions were seen in only 27.3% of 1,882 cancers without *8q* gain but in 57.4% of 122 tumors with *8q* gain ($P < 0.0001$). Among cancers treated with radical prostatectomy, *8p* deletions ($P = 0.003$) and *8q* gains ($P = 0.02$) were associated with biochemical tumor recurrence. However, multivariate analysis (including prostate-specific antigen, pT/pN stage, Gleason score, and surgical margin status) did not reveal any statistically independent effect of 8p or 8q alterations on biochemical tumor recurrence.

Conclusions: *8p* deletions and *8q* gains are relatively rare in early stage prostate cancer but often develop during tumor progression. The prognostic effect does not seem to be strong enough to warrant clinical application. *Clin Cancer Res; 16(1); 56–64. ©2010 AACR.*

Prostate cancer is the most frequently diagnosed malignancy and the second most frequent cause of cancer-related death among western males (1).

Currently, clinical prediction tools rely solely on clinical (clinical stage), serologic (prostate-specific antigen), and histologic variables (Gleason grading; ref. 2).

Because the development and progression of cancer is driven by molecular alterations, the analysis of molecular features may eventually allow better prediction of the behavior of individual cancers.

Authors' Affiliations: [1]Institute of Pathology, [2]Martini-Clinic, Prostate Cancer Center, [3]Departments of Gynecology, [4]Oncology and Hematology, [5]Thoraco-Visceral Surgery, and [6]Urology, University Medical Center, Hamburg, Germany

Note: T. El Gammal, M. Brüchmann, and J. Zustin contributed equally to this work.

Corresponding Author: Thorsten Schlomm, Martini-Clinic, Prostate Cancer Center, University Medical Center Hamburg-Eppendorf, Martinistr. 52, Hamburg 20246, Germany. Phone: 49-40-7410-51314; Fax: 49-40-7410-51323; E-mail: tschlomm@uke.de.

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Alterations on the DNA level would be especially suitable as prognostic markers. DNA alterations have the advantage of being less vulnerable to perioperative and postoperative ischemia or fixation procedures than RNA or proteins (3, 4). Deletions of *8p* and gains of *8q* are among the most frequent genomic alterations in prostate cancer. Several studies have proposed a relationship between *8p−/8q+* with adverse histopathologic findings (5–11). Studies suggesting associations with prostate-specific antigen progression, however, were limited to <100 cancers (12–14). Whereas considerable evidence points toward a role of chromosome 8 changes in prostate cancer biology, it is also evident that much larger patient cohorts are now needed to further clarify the clinical utility of *8p* and *8q* analysis. Our group had previously manufactured a tissue microarray (TMA) consisting of tumors from >3,000 prostate cancer patients (15–19). This TMA allows a rapid parallel *in situ* analysis of molecular features in a contemporary and homogeneously treated patient cohort. The examination of *8p/8q* changes on this TMA points toward a significant relevance of *8p* deletion and *8q* gains in different stages of prostate cancer progression.

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Translational Relevance

The clinical and biological significance of deletions of *8p* and gains of *8q* in prostate cancers are widely unknown. In this study, a set of almost 2,000 prostate cancers with attached longtime follow-up was successfully analyzed by fluorescence *in situ* hybridization for chromosome 8 alterations. *8p* deletions could already be frequently detected in early cancers, and the frequency did not significantly increase from localized to metastatic tumors, suggesting an early and highly conserved event in tumor initiation and progression. In contrast, the frequency of *8q* gains increased sharply between localized primary tumors, and advanced metastatic and hormone refractory tumors, raising the possibility that at least one *8q* gene is relevant for progression to the deadly end stages of prostate cancer. Despite the biological importance of these results, the prognostic effect of the chromosome 8 status is not strong enough to add significant prognostic information to the established clinicopathologic variables.

Materials and Methods

Patients. Radical prostatectomy specimens were available from 3,261 patients consecutively treated at the Department of Urology, University Medical Center Hamburg-Eppendorf, between 1992 and 2005 (Table 1). The study cohort represents a referred "early detection" population. Follow-up data were available for 2,385 patients, ranging from 1 to 144 mo (mean, 34 mo). None of the patients received neoadjuvant or adjuvant therapy. Additional (salvage) therapy was only initiated in case of a biochemical relapse. In all patients, prostate-specific antigen values were measured quarterly in the 1st year, followed by biannual measurements in the 2nd year and annual measurements after the 3rd year after surgery. Recurrence was defined as a postoperative prostate-specific antigen of 0.1 ng/mL and higher. The first prostate-specific antigen value ≥0.1 ng/mL was used to define the time of recurrence. Patients without evidence of tumor recurrence were censored at last follow-up. All prostatectomy specimens were analyzed according to a standard procedure. All prostates were completely paraffin embedded, including whole-mount sections, as previously described (20). All H&E-stained histologic sections from all prostatectomy specimens were reviewed, and the index tumors, defined as the largest tumor focus and/or the focus with the worst Gleason pattern, were marked on the slides. One 0.6-mm tissue core was punched out from the index tumors of each case and transferred in a TMA format as described (21). The 3,261 cores were distributed among seven TMA blocks, each containing 129 to 522 tumor samples. Each TMA block also contained various control tissues, including normal prostate tissue. In addition, 23 lymph node metastases, 27 bone metastases, and 22 hormone refractory cancers were analyzed on a separate progression TMA (tissues were not matched to the radical prostatectomy specimens). Hormone refractory prostate cancer was defined as: serum castration levels of testosterone, three consecutive increases in the prostatic-specific antigen resulting in two 50% increases over the nadir, antiandrogen withdrawal for at least 4 wk, prostate-specific antigen progression despite secondary hormonal manipulations, or progression of osseous or soft tissue lesions (22).

Immunohistochemistry. Freshly cut TMA sections were analyzed in 1 d in one experiment for each antibody. Slides were immunostained for high molecular weight cytokeratins to assure the presence of cancer cells in the TMA spots.

Table 1. Descriptives of the entire study cohort of 1,882 patients who were treated with radical prostatectomy for clinically localized prostate cancer

Variable	No. of patients
PSA (ng/mL)	
Mean (median)	10.0 (6.9)
Range	0.1-137.0
pT stage	
pT_2	1,239 (65.8%)
pT_{3a}	379 (20.1%)
pT_{3b}	237 (12.6%)
pT_4	27 (1.4%)
Prostatectomy Gleason score	
≤6	832 (44.2%)
3+4	827 (43.9%)
4+3	194 (10.3%)
≥8	29 (1.5%)
pN stage	
pN_0	960 (51.0%)
pN_{1-3}	59 (3.1%)
pN_x	863 (45.9%)
Surgical margin status	
Negative	387 (20.6%)
Positive	1,495 (79.4%)
Chromosome 8p alterations	
Normal	1,299 (69.0%)
Loss	583 (31.0%)
Chromosome 8q alterations	
Normal	1,760 (93.5%)
Gain or amplification	122 (6.5%)
Combined alteration	
None	1,247 (66.3%)
One "region"	565 (30.0%)
Both "regions"	70 (3.7%)
Follow-up of censored patients (mo)	
Mean (median)	36.8 (34.7)
Range	1.0-133.0
Overall no. of biochemical recurrences	328 (17.4%)
5-year biochemical recurrence–free survival	69.6%

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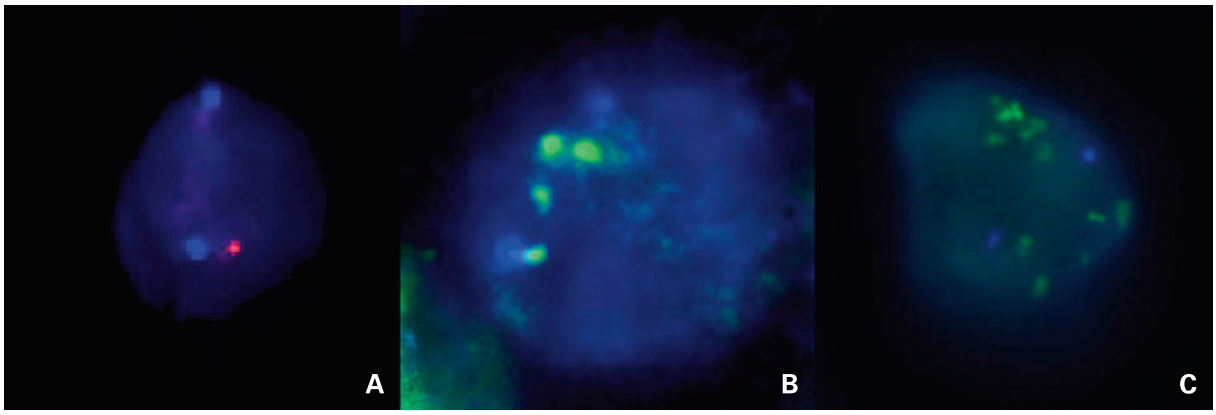

Fig. 1. Examples for *8p* deletion and *8q* gains in prostate cancer cells. *A*, cell nucleus (blue) showing two copies of centromere 8 (dark blue signals) but only one copy of the LPL gene (red signal) located at chromosomal band *8p21*, indicating loss of chromosome *8p*. *B*, cell nucleus with 5-6 copies of *MYC* located at chromosomal band *8q24* and two blue centromere 8 signals indicating gain of chromosome *8q*. *C*, high-level amplification of *MYC* (20-22 green signals) was found in a single case only.

For this purpose, the antibody 34βE12 (clone MA903; Dako; 1:12.5) was used for basal cell detection after boiling the sections in an autoclave in citrate buffer (pH7.8). Tissues from cancer-free prostates served as negative and positive controls on each TMA section. Only tissue samples with distinct loss of basal cells (proven prostate cancers) were used for *8p*/*8q* analysis.

Fluorescence* in situ *hybridization. A 4-μm TMA section was used for multicolor fluorescence *in situ* hybridization (FISH). For proteolytic slide pretreatment, a commercial kit was used (paraffin pretreatment reagent kit; Vysis, Downers Grove, IL). The multicolor FISH probe set consisted of a Spectrum Orange–labeled *LPL* probe (8p21), a Spectrum Green–labeled *MYC* probe (8q24), and an aqua blue–stained centromere 8 probe (ProVysion; Vysis). Before hybridization, TMA sections were deparaffinized, air-dried, and dehydrated in 70%, 85%, and 100% ethanol, followed by denaturation for 5 min at 74°C in 70% formamide 2x SSC solution. After overnight hybridization at 37°C in a humidified chamber, slides were washed and counterstained with 0.2 μmol/L 4′,6-diamidino-2-phenylindole in an antifade solution. Each tumor spot was carefully visually evaluated, and the predominant score was recorded for each FISH probe. For example, this procedure always resulted in a score of 2 centromere 8, 2 *MYC*, and 2 LPL for normal tissues. For subsequent statistical analyses, groups were defined as follows: an *8q* gain was defined as the presence of at least one more copy of *MYC* compared with centromere 8, and an *8p* deletion was defined as the presence of at least one less copy of *LPL* compared with centromere 8. Examples of tumors with chromosome 8 alterations are shown in Fig. 1. These definitions were previously used in a multitude of studies investigating gene copy numbers by FISH (23, 24).

Statistics. Independent *t* test and χ^2 test were used for comparison of means and proportions between groups, respectively. Kaplan-Meier plots were used to graphically illustrate the biochemical recurrence–free survival estimates for patients with versus those without chromosomal alterations. Differences between groups were assessed with the use of the log-rank test. Univariate and multivariate Cox regression models tested the effect of chromosome *8p* (loss versus normal) and/or *8q* (gain/amplification versus normal) alterations on biochemical recurrence. Covariates consisted of prostate-specific antigen, pT stage, pN stage, prostatectomy Gleason score, and surgical margin status. The univariate predictive accuracy of biochemical recurrence was assessed for each of the predictor variables. In all analyses, predictive accuracy was quantified according to the method described by Harrell et al. and was expressed as a percentage from 50% to 100%, of which 50% is equivalent to a flip of a coin and 100% represents perfect discrimination (25, 26). Subsequently, the combined predictive accuracy of all standard variables (prostate-specific antigen, pT stage, pN stage, prostatectomy Gleason score, and surgical margin status) was quantified and termed the accuracy of the base multivariate model. The change in multivariate predictive accuracy related to the inclusion of chromosome *8p* and/or *8q* alterations was then tested. To reduce overfit bias, all univariate and multivariate models were subjected to 200 bootstrap resamples. All statistical tests were done with the use of S-PLUS Professional version 1 (Mathsoft, Seattle, WA). All tests were two-sided, with a significance level set at 0.05.

Results

Technical issues. For *8p*/*8q* FISH analysis, 1,164 of 3,261 cancers (35.7%) were noninformative due to a complete lack of tissue samples or absence of unequivocal cancer tissue in the corresponding TMA section immunostained with 34BE12. A total of 2,097 cancers were successfully

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Table 2. Correlation between clinical and histopathologic findings according to 8p and 8q status

	Analyzable tumors (*n*)	8p deletion	*P*	8q gain	*P*
pT stage					
Organ-confined disease (pT_2)	1,239	323 (26.1%)	<0.0001	60 (4.8%)	<0.0001
Extraprostatic extension (pT_{3a})	379	146 (38.5%)		37 (9.8%)	
Seminal vesicle invasion (pT_{3b})	237	103 (43.5%)		21 (8.9%)	
pT_4 disease	27	11 (40.7%)		4 (14.8%)	
pN stage*					
pN_0	960	346 (36.0%)	0.2	77 (8.0%)	0.6
pN_1	59	26 (44.1%)		6 (10.2%)	
Prostatectomy Gleason score					
≤6	832	204 (24.5%)	<0.0001	29 (3.5%)	<0.0001
3+4	827	274 (33.1%)		63 (7.6%)	
4+3	194	91 (46.9%)		26 (13.4%)	
≥8	29	14 (48.3%)		4 (13.8%)	
Surgical margin status					
Negative	1,495	443 (29.6%)	0.01	95 (6.4%)	0.7
Positive	387	140 (36.2%)		27 (7.0%)	

*Only patients who underwent a formal lymph node dissection were included.

analyzed by FISH for *8p* deletions and *8q* gains. From 215 of these patients, complete follow-up data were not available, resulting in 1,882 analyzable primary cases with follow-up data (Table 1), and an additional set of 72 metastases and hormone refractory cancers. The 1,379 samples that were excluded from analyses due to technical issues or missing clinical data did not differ in a statistically significant fashion from the samples included in the analyses (data not shown). In consequence, the exclusion of those patients did not result in selection bias.

8p deletions. *8p* deletions according to our definition were found in 616 (31.5%) of the 1,954 interpretable cancers. There was a significant association of *8p* deletions with unfavorable tumor phenotype. *8p* deletions were seen in 26.1% of 1,239 pT_2 cancers and in 24.5% of 832 cancers with a Gleason score ≤3+3. The frequency of deletions increased continuously with advancing stage, and higher Gleason grade ($P < 0.0001$ each) and surgical margin status ($P = 0.01$; Table 2), but was unrelated to nodal status. Although there was a slight increase in 8p deletions from pT_3 primary tumors to bone metastases or hormone refractory cancer, these differences did not reach statistical significance. Among primary tumors treated with radical prostatectomy, *8p* deletions were significantly associated with poor prognosis ($P = 0.003$; Fig. 2A).

8q gains. *8q* gains were typically low-level with up to seven MYC gene copies. Only one of 1,954 interpretable cases showed a high-level amplification with >20 gene copies (Fig. 1B). *8q* gains were markedly less frequent in untreated primary tumors (<8%) than *8p* deletions (>30%; Table 2). In our radical prostatectomy samples, the increase in *8q* gains with unfavorable tumor phenotype was relatively moderate. *8q* gains increased from 4.8% in pT_2 cancers to 8.9% in pT_{3b} cancers ($P < 0.0001$) and from 3.5% in Gleason ≤3+3 cancers to 13.8% in Gleason ≥4+4 carcinomas ($P < 0.0001$). However, the frequency of *8q* gains increased sharply from primary cancers to metastatic [both nodal (34.8%) and bone (33.3%)] and hormone refractory cancer (59.9%; $P < 0.0001$ each; Fig. 3). *8q* gains were associated with poor survival in primary cancers ($P = 0.02$; Fig. 2B). *8q* gains and *8p* deletions often co-occurred. *8p* deletions were seen in only 27.3% of 1,882 primary prostate cancers without *8q* gain but in 57.4% of 122 primary tumors with *8q* gain ($P < 0.0001$). However, a separate survival analysis analyzing tumors with both alterations versus only one alteration showed no significant prognostic difference ($P = 0.1$; Fig. 2C).

Multivariate analyses. In multivariate models, all tested established predictors (prostate-specific antigen, pT stage, pN stage, Gleason score, and surgical margin status) reached independent predictor status of biochemical recurrence (all adjusted $P < 0.0001$; Table 3). Conversely, all tested *8p* and *8q* alterations (*8p* normal versus loss, *8q* normal versus gain/amplification, and number of alterations: 0 versus 1 versus 2) failed to independently predict biochemical recurrence (all adjusted $P \geq 0.03$). The multivariate accuracy of the base model, which relied on all established variables, was 80.6% accurate in predicting biochemical recurrence. The addition of *8p* and *8q* status to the base model could not improve the combined multivariate predictive accuracy (80.6% versus 80.5%). This finding indicates that the addition of chromosome 8 alterations neither independently predicts biochemical recurrence nor increases the combined predictive accuracy of established biochemical recurrence predictors (Table 3).

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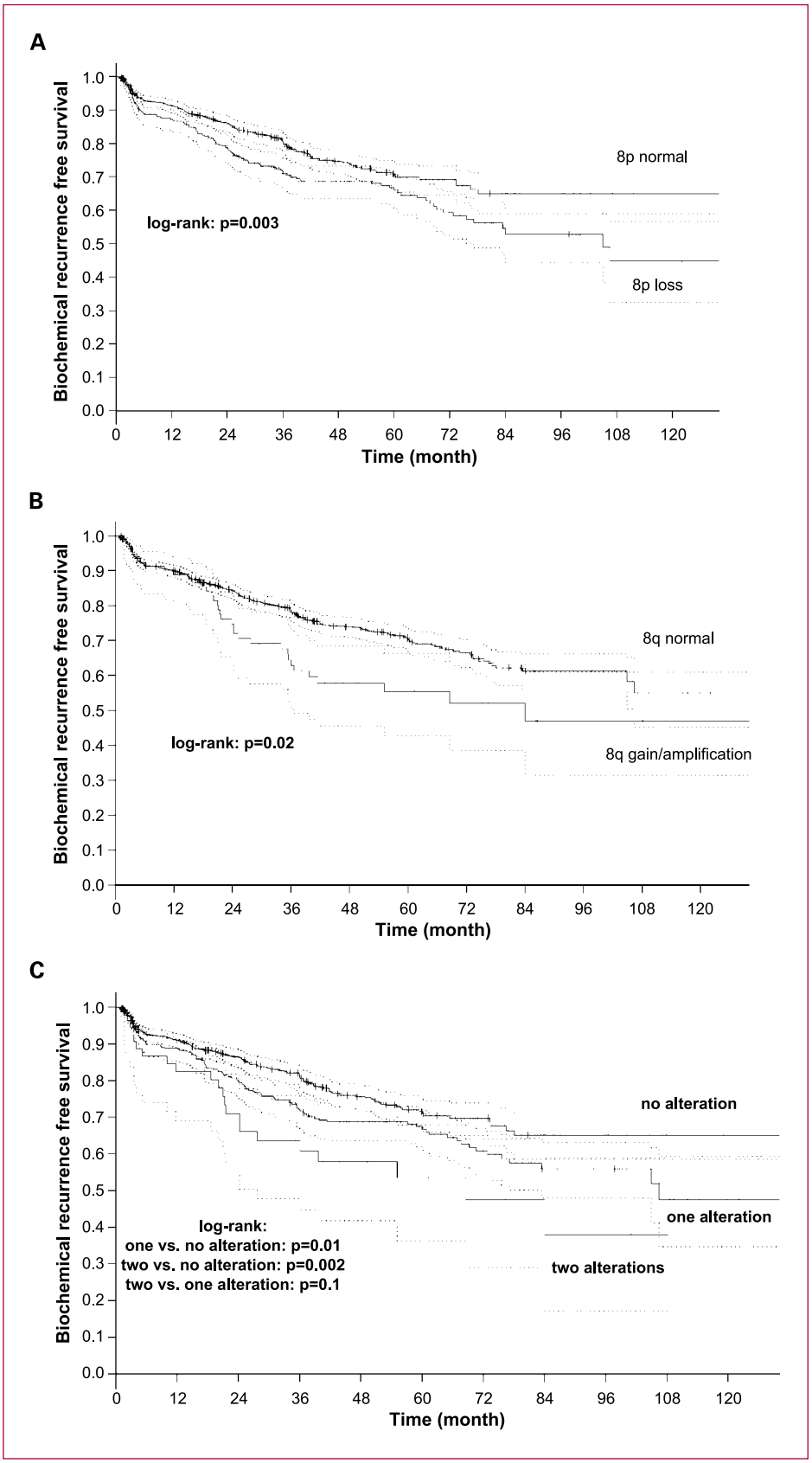

Fig. 2. Kaplan-Meier plots illustrating the effect of chromosome 8 alterations on the risk of biochemical recurrence after radical prostatectomy. *A*, *8p* losses. *B*, *8q* gains/amplifications. *C*, combined *8p*/*8q* alterations. Dots indicate the 95% confidence intervals.

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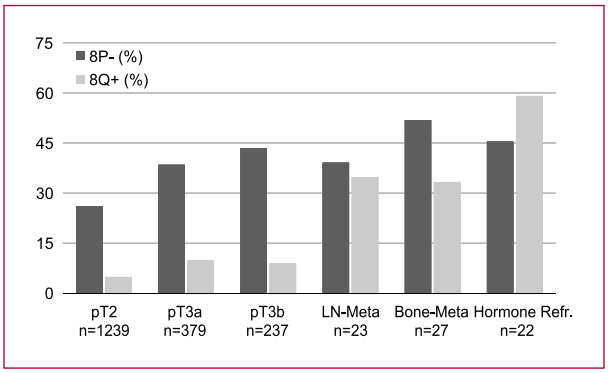

Fig. 3. Chromosome 8 alterations and tumor-stage.

Discussion

The frequency of *8p* deletions and *8q* gains found in this study is substantially lower than in the majority of previous FISH studies, in which frequencies between 50.0% (5, 9) and 74% had been reported for *8p* deletions (5, 6, 8, 12, 14, 27–29), and between 6.1% (30) and 73.3% (13) for *8q* gains (5, 7, 9, 12, 29, 30). This seeming discrepancy is mainly caused by the different aims and approaches of the different studies. The ProVysion test used in the present study has initially been introduced with the purpose of facilitating prostate cancer diagnosis with the use of *8p22* (LPL) and *8q24* (*MYC*) probes as surrogate markers for large *8p* losses and *8q* gains, which were often observed in studies that use classical comparative genomic hybridization (32, 33). The threshold used in early studies was therefore selected to detect significant abnormalities of *8p* and *8q* copy numbers in cancer compared with non-neoplastic tissues. Accordingly, thresholds were usually based on scores obtained in normal tissue. A deviation of +/− 2 or 3 SDs from the findings in normal tissues was used to define *8p*/*8q* alterations by other authors (9, 14, 31). Under such criteria, cancers with a significant chromosomal instability leading to a certain number of nonclonal chromosomal alterations, including *8q+* or *8p−*, will be identified as aberrant. It is also noteworthy that some of the differences in FISH findings between normal and cancer tissue can always be explained by

Table 3. Univariate and multivariate Cox regression models testing the effect of several predictors on the risk of biochemical recurrence after radical prostatectomy

Variable	Univariate		Multivariate	
	HR, *P*	Accuracy	HR, *P*	HR, *P*
PSA	1.02, $P < 0.0001$	64.6%	1.01, $P = 0.01$	1.01, $P = 0.001$
pT stage	$P < 0.0001$	—	$P < 0.0001$	$P < 0.0001$
pT_{3a} vs pT_2	3.38, $P < 0.0001$	74.2%	1.97, $P < 0.0001$	1.98, $P < 0.0001$
pT_{3b} vs pT_2	8.60, $P < 0.0001$	—	3.24, $P < 0.0001$	3.26, $P < 0.0001$
pT_4 vs pT_2	13.67, $P < 0.0001$	—	4.04, $P < 0.0001$	4.14, $P < 0.0001$
pN stage	$P < 0.0001$	—	$P < 0.0001$	$P < 0.000$
pN_1 vs pN_0	6.52, $P < 0.0001$	56.5%	1.83, $P = 0.001$	1.83, $P = 0.001$
pN_x vs pN_0	0.87, $P = 0.4$	—	1.60, $P = 0.004$	1.60, $P = 0.04$
Prostatectomy Gleason score	$P < 0.0001$	—	$P < 0.0001$	$P < 0.0001$
3+4 vs ≤6	4.18, $P < 0.0001$	74.2%	2.62, $P < 0.0001$	2.62, $P < 0.0001$
4+3 vs ≤6	15.89, $P < 0.0001$	—	6.99, $P < 0.0001$	6.91, $P < 0.0001$
≥8 vs ≤6	22.18, $P < 0.0001$	—	7.20, $P < 0.0001$	7.25, $P < 0.0001$
Surgical margin status				
Positive vs negative	2.68, $P < 0.0001$	60.4%	1.68, $P < 0.0001$	1.68, $P < 0.0001$
8p status				
Normal vs loss	1.39, $P = 0.003$	54.4%	1.04, $P = 0.8$	—
8q status				
Normal vs gain/amplified	1.55, $P = 0.02$	51.6%	1.24, $P = 0.3$	—
8p + *8q* status	$P = 0.001$			$P = 0.4$
One alteration vs both normal	1.35, $P = 0.01$	54.7%	—	1.02, $P = 0.9$
Two alterations vs both normal	1.98, $P = 0.002$			1.37, $P = 0.2$

Accuracy	%
Accuracy of the base model (PSA, pT stage, pN stage, prostatectomy Gleason score, and surgical margin status)	80.6%
Accuracy of the base model + *8p* status	80.5%
Accuracy of the base model + *8q* status	80.5%
Accuracy of the base model + *8p* and *8q* status	80.5%

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the size difference between normal and cancerous cells. The larger diameter of cancer cells leads to a higher probability of losing signals because of nuclear truncation. This will more greatly affect the smaller locus specific signals (*8p*/*8q*) than the relatively large centromere 8 reference probes. Furthermore, most studies analyzing chromosome 8 alterations were done between 1995 and 2002 on historical patient collectives containing a high proportion of high-stage and advanced tumors (10, 32–37). Our patient cohort consists of >60% organ-confined (pT_2) tumors.

The present study was undertaken to determine the potential prognostic/clinical significance of *8p* deletions and *8q* gains. The scoring criteria were therefore selected to identify cancers with unequivocal *8p*– or *8q*+ in clonally expanding cells. Only tumors with *8p* deletions or *8q* gains in virtually all cells will potentially be driven by inactivation of one or several genes on *8p* or overexpression of *8q* gene (s). Our notion that the frequencies of *8p* deletions and *8q* gains found in this project may reflect the true fraction of cancers having a "dominant" proportion of such cells is supported by comparable numbers found by methods analyzing "average copy numbers" of genomic material, such as loss of heterozygosity or comparative genomic hybridization. Studies analyzing comparable tumor populations by comparative genomic hybridization have reported between 17.6% and 32% *8p* deletions for primary tumors (32, 38–40), and between 48% and 73% for advanced tumors (41–44). In a recent meta-analysis, chromosome *8p* was the most deleted region in the genome (45). The peak frequency for *8p* deletions (peak loss, *8p21.3*) in primary tumors was 34.09%, a figure that almost exactly matches our frequency of 31.55% observed by FISH.

Our rather low results for *8q* gains in early stage tumors are supported by some comparative genomic hybridization analyses with comparable frequencies between 6% and 9% (32, 38–40). From the data, it seems that these studies had preferably included early stage tumors. There seems to be a significant difference between primary tumors and advanced tumors in gain of *8q*. Due to the fact that advanced tumors (locally advanced to metastatic tumors) were overrepresented in the majority of comparative genomic hybridization studies, the numbers for *8q* gains found by comparative genomic hybridization from 29.6% to 47.8% (41–46) were also comparable with our results (34. 8%/33.3%/59.9% in lymph node metastatic/bone metastatic/hormone refractory cancers).

Our data show a significant increase in both *8p* deletions and *8q* gains with tumor stage and grade. However, the relationship with tumor phenotype varied sharply between these alterations. *8p* deletions were already frequent in early stage cancers and increased only mildly from small (pT_2) to more extended cancers (pT_3), and metastatic or hormone refractory disease (Fig. 3).

This substantial difference between localized and metastatic prostate cancer suggests a potential role of one or several *8q* genes for metastasis. Such a scenario would also be consistent with a markedly worse prognosis found for these few localized prostate cancers having *8q* gains. An involvement of *8q* genes in the metastatic cascade had also been suggested for other cancer types (47).

It was previously discussed whether our approach of using just one 0.6-mm tissue core per cancer might be sufficiently representative of heterogeneous tumors, such as prostate cancer (48). The use at least four samples per tumor has been suggested because better associations with large-section results could be achieved by this method. We are uncertain about this concept because of two reasons. First, cancer is usually found in many of the blocks made from one radical prostatectomy specimen (in our laboratory, 7-8 blocks on average contain cancer). To exclude the effect of cancer heterogeneity on the study results, it seems quite insufficient to optimize the analysis of one of these blocks. Second, TMA analysis of multiple tissue cores from one tissue block undoubtedly leads to more "positive" cases. This includes true positive cases as well as false positive cases due to interpretation problems and artifacts. For example, it has been estimated that 10% to 20% of *HER2* results may suffer from technical and interpretation errors in breast cancer (49, 50). In TMA studies, however, such artifacts do not exert significant influence on the study outcome because of the high number of samples typically included in this kind of analysis. This is best illustrated by the fact that virtually all studies analyzing clinical markers in large TMAs were able to fully reproduce the known prognostic effect (15, 18, 51). Whereas many studies show that more TMA cores per block lead to better association with the result obtained on the corresponding large section (52), there are no studies (to our knowledge) showing an improved relationship with prognosis or tumor phenotype when more cores are used in studies with sufficiently large TMAs. Importantly, there was even one large study in which p53 was found to be a highly significant prognostic marker in four different one-core TMAs of >500 breast cancers. However, although the corresponding large section showed 80% more positive cases, the large-section results were not associated with prognosis. It was concluded that false or irrelevant p53 staining was overinterpreted in the large-section analysis (52).

The aim of this project was to determine whether or not *8p*– or *8q*+ are related to prostate cancer progression and prognosis. The suitability of our TMA for this purpose can hardly be challenged because highly significant associations of chromosome 8 alterations with tumor phenotype and prognosis were found. The data show that *8p* deletions and *8q* gains are related to prostate cancer progression. However, the *8p*/*8q* status does not add significant information to increase the predictive accuracy of the established clinicopathologic variables. It is possible, however, that the true prevalence of heterogeneous chromosome 8 alterations is higher than the figures identified in this study. To determine the frequency of small heterogeneous *8p* deletions and *8q* gains, it will be necessary to analyze all available tumor blocks from a representative number of cancers.

In summary, the results of this study show that *8p* deletions and *8q* gains are important elements in prostate

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cancer progression. Studies are needed to identify the genes driving these alterations. The striking association of *8q* gains with metastasis and hormone refractory disease raises the possibility that at least one *8q* gene is relevant for progression to the deadly end stages of prostate cancer.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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Alexander T. El Gammal, Michael Brüchmann, Jozef Zustin, et al.

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