Homozygous and Frequent Deletion of Proximal 8p Sequences in Human Prostate Cancers: Identification of a Potential Tumor Suppressor Gene Site

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By using tissue microdissection and polymerase chain reaction (PCR) techniques, we examined 85 prostate tumors that were paired with normal tissues from the same patients for allelic loss at 26 highly polymorphic microsatellite sequences, 21 spanning 8p and 5 localized to 8q. Sixty-four tumors (75%) demonstrated loss of at least one 8p locus. Separate distal and proximal regions of deletion were observed as well as an intervening, staggered breakpoint. A novel region of homozygous deletion of sequences at the D8S87 locus was detected both by multiplex PCR and by fluorescence in situ hybridization within this breakpoint region. These data suggest that a tumor-suppressor gene mapping to proximal 8p is deleted frequently and is likely to be important for tumorigenesis in prostate tumors. *Genes Chromosomes Cancer 23:255–262,1998.*

INTRODUCTION

It is likely that many genetic and epigenetic events are involved in tumorigenesis. In particular, recent cytogenetic and molecular studies have suggested that deletion or rearrangement of sequences that map to the short arm of chromosome 8 may be permissive for tumorigenesis in several organ systems. In prostate carcinoma, early work by Bergerheim et al. (1993) utilizing Southern blot techniques observed high frequencies of deletion of distal 8p sequences in human prostate tumors, suggesting that a breakpoint existed between the *NEFL* locus at 8p21 and the *PLAT* locus at 8p21–8p11. Subsequent work by Bova et al. (1993) and our laboratory (Wolman et al., 1992; Macoska et al., 1993) identified a major region of deletion mapping to 8p22 in human prostate cancer (PCa), which is characterized by a homozygous deletion mapping within the 8p22–p23 region in a prostate tumor (Bova et al., 1996). The use of polymerase chain reaction (PCR) and fluorescence in situ hybridization (FISH) techniques in our laboratory and by others confirmed frequent heterozygous loss of distal 8p sequences (Macoska et al., 1994; MacGrogan et al., 1994; Sakr et al., 1994) and showed that this region of deletion could be quite extensive, including the entire 8p chromosome arm or large portions of 8p extending from 8pter to 8p11–12 (MacGrogan et al., 1994; Trapman et al., 1994). We later reported interstitial deletions of distal sequences mapping to 8p22 or 8p21 or proximal loci mapping to 8p11–12 in human prostate tumors (Macoska et al., 1995). Other laboratories reported deletional domains encompassing or including 8p22 and 8p21-p12 (Vocke et al., 1996), interstitial losses and homozygous deletions at 8p21 and 8p22 (Kagan et al., 1995; Suzuki et al., 1995), interstitial losses of two separate proximal deletional domains (Crundwell et al., 1996), and isolation of a potential tumor suppressor gene mapping to 8p21.3–p22 (Fujiwara et al., 1995). Recent studies have also mapped a putative prostate metastasis suppressor gene to the 8p21-p12 region (Nihei et al., 1996).

Taken together, these studies suggest that there are at least two large 8p sequence domains that are deleted in prostate, bladder, and other cancers: distal sequences encompassing 8pter-p21 (sometimes divided further into smaller, separate deletional domains) and proximal sequences encompassing 8p11–p12. In this report, we present data that demonstrate frequent and sometimes homozygous deletion of specific sequences mapping to proximal 8p. This information should facilitate the isolation and cloning of one or more putative proximal 8p tumor-suppressor genes that are inactivated in a variety of human cancers.

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MATERIALS AND METHODS

Tissue Characterization

Prostate tissue was obtained after radical prostatectomy from 85 patients diagnosed with PCa. After an initial pathologic evaluation of radical prostatectomy tissue, presumed malignant and normal tissue was snap frozen in liquid nitrogen and stored at −70°C. Tumor specimens comprising areas of at least 70% malignant cells and nontumor specimens comprising normal or hyperplastic epithelium were serially sectioned at 4 microns. One section was stained with hematoxylin/eosin for histologic evaluation, and discrete areas of benign or malignant glands were then excised from adjacent nonstained sections. DNA was extracted as described previously (Macoska et al., 1993, 1994, 1995; Sakr et al., 1994). Serum prostate-specific antigen (PSA) values were obtained within a 1-week period prior to surgery.

Analysis of DNA for Allelic Loss

PCR was utilized for amplification of 26 sequences containing highly polymorphic microsatellite repeat markers at loci of interest on chromosome 8:21 loci that map to 8p and 5 that map to proximal 8q. The linkage order of these markers has been reported as pter-D8S549-D8S1715-D8S1116/D8S1739-D8S1725-*NEFL*-D8S137-D8S1423-D8S540/D8S1125/D8S259/D8S513/D8S1105/D8S505-D8S535-D8S87-D8S255-D8S1118-D8S1121-AFMa295zd5-D8S1104-D8S531-D8S519-D8S1098-D8S538-D8S589-qter (where / indicates markers of uncertain relative order; see Fig. 1). Microsatellite-specific primer sequences mapping near the *WRN* locus at the D8S1105 locus were as reported (Ye et al., 1995). Cytogenetic localizations were available for the following markers: 8pter-p23.1, D8S549; 8p21, *NEFL*; 8p12, D8S87; and 8q11.2-q12, D8S1104 (as reported by the Human Genome Data Base; GDB; http://gdbwww.gdb.org). Primer sequences, additional linkage and contig information, and genetic mapping information were obtained from public databases maintained by the GDB, the Center for Genome Research at the Whitehead Institute for Biomedical Research (http://www-genome.wi.mit.edu/), the Cooperative Human Linkage Center (CHLC; http://www.chlc.org/), and the National Center for Biotechnology Information (http://www.ncbi.nlm.nih.gov/), as accessed through the Internet.

PCR reactions were performed as described previously (Trybus et al., 1996). Aliquots of each reaction were electrophoresed on 6% acrylamide/7 M urea sequencing gels, and the gels were autoradiographed. Allelic loss was scored when the ratio of allelic signal intensities in tumor tissue was ≤ 50% of that for the same alleles in normal tissue from the same heterozygous patient by two independent observers (M.A.P. or T.M.T. and J.A.M.). Statistical analysis was performed by χ^2 test, with P values < 0.05 considered statistically significant.

Instances of putative homozygous loss were investigated further by multiplex PCR analysis, in which two primer sets were introduced simultaneously into the same reaction mixtures. The failure to produce an amplification product from one of the two amplicons in a multiplex reaction was interpreted as homozygous loss of the sequence that failed to amplify.

Fluorescence In Situ Hybridization

Slides were prepared from frozen tissues as described previously (Macoska et al., 1994). A clone, 303f13, mapping to the D8S87 locus was isolated by PCR amplification of a microsatellite marker specific for that region from a human BAC DNA library (Research Genetics, Inc., Huntsville, AL). Subsequent analysis confirmed that BAC 303f13 contains sequences specific for D8S87 (cytogenetically localized to 8p12) and WI-4330 (mapped to chromosome 8) and produces no more than two discrete fluorescent signals when it is labeled and hybridized to cut sections of normal prostatic epithelium (see Table 1). These findings are consistent with an exclusive chromosome 8 localization for this BAC. One microgram of BAC 303f13 was labeled via nick translation with 20 μM fluorescein-11-dUTP (FluoroGreen; Amersham, Buckinghamshire, United Kingdom). Unincorporated nucleotide was eliminated by passage through Sephadex G-25. The labeled DNA was digested with EcoRI and ethanol precipitated in the presence of Cot-1 (10 μg) DNA, yeast tRNA (25 μg), and sonicated salmon testes DNA (25 μg). The probe was then resuspended in 10 μl formamide and combined with an equal volume of hybridization solution (to final concentrations of 25% dextran sulfate, 2.5 μg/ml bovine serum albumin, 2.5 × standard saline citrate; SSC). One microliter of rhodamine-labeled chromosome 8 pericentromeric probe DNA (CEP8 SpectrumOrange; Vysis, Downers Grove, IL) was added to the hybridization mixture and applied to the prepared slide, which was coverslipped and sealed. The slide was incubated at 90°C for 10 minutes and was then incubated at 37°C overnight. Unbound probe was removed by washing the slide

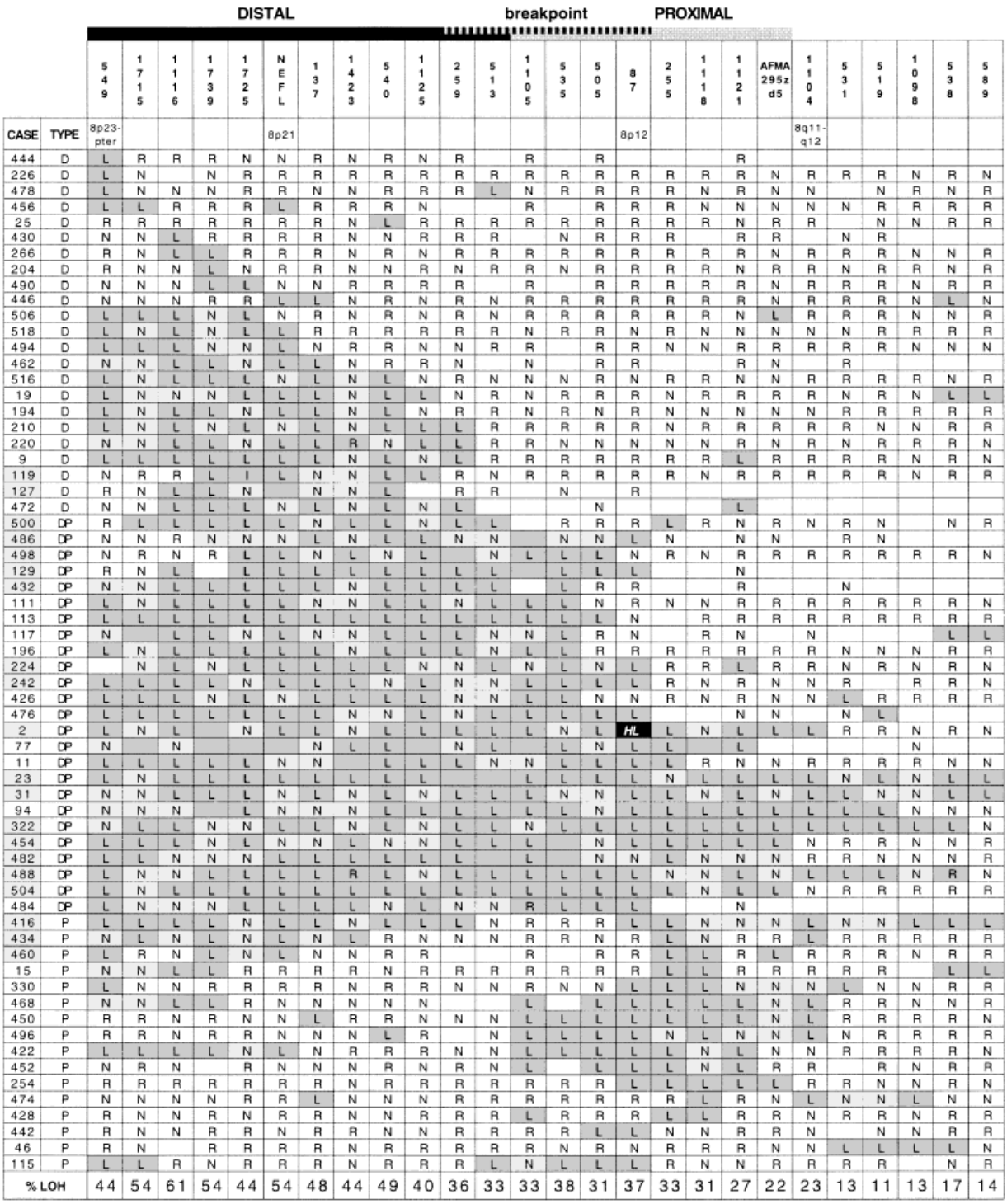

Figure 1. Deletion profile of 64 prostate tumors with allelic loss of 8p sequences. Loci are shown at the top of the figure and are oriented from 8pter on the left to 8cen on the right and refer to ''D8'' designations (with the exceptions of the *NEFL* and AFM295zd5 loci). Prostate tumors are refered to by case number on the y-axis. Type refers to major region of deletion, D (distal), P (proximal), or DP (distal + proximal). Allelic loss patterns are indicated as follows: R, both alleles retained (white boxes); N, not informative (light gray boxes); and L, loss of one allele (dark gray boxes). Extended light and dark gray shaded areas show probable contiguous regions of deletion. The black box designated ''HL'' shows a region of homozygous allelic loss. The extents of the distal (black bar) and proximal (gray bar) regions of deletion are indicated at the top, as is the breakpoint (black and gray hatched bar). Allelic loss frequencies for individual loci (out of a total of 85 tumors) are shown at the bottom. Specific cases discussed in the text are highlighted.

TABLE 1. Fluorescence In Situ Hybridization Analysis of Homozygous Deletion at D8S87

8p,8c Signal counts	Normal tissue nuclei (%)	Tumor tissue nuclei (%)
Case 2		
1,1	9 (5)	8 (4)
1,2	26 (13)	61 (30)
2,2	123 (62)	19 (10)
0,2	15 (7)	97 (49)
Other	27 (13)	15 (7)
Total	(100)	(100)
Total 8p loss	50 (25)	166 (83)
Case 512		
1,1	10 (5)	2 (1)
1,2	48 (24)	46 (23)
2,2	108 (54)	108 (54)
0,2	18 (9)	27 (13)
Other	16 (8)	17 (9)
Total	(100)	(100)
Total 8p loss	76 (38)	75 (38)

in 2 × SSC for 30 minutes each successively at room temperature, at 37°C, and at 45°C, then at 70°C for 5 minutes. The slide was stored at 4°C in 1 × phosphate-buffered detergent (0.1M Na_2HPO_4, 0.1M NaH_2PO_4, 0.5% NP-40) (PBD) prior to counterstaining and visualization.

Nuclei were counterstained with DAPI/antifade, and slides were visualized using a mercury light source and appropriate filters (Chroma Optical, Brattelboro, VT) on a photomicroscope (Zeiss Axiophot, Thornwood, NY). The x,y coordinates of microscope fields at × 1,000 within areas of appropriate histology of normal and tumor tissues were noted, and nuclei of 200 epithelial cells were enumerated for the BAC and CEP8 probes. Careful attention was given to counting only tissue areas of appropriate histology with good nuclear morphology. Two observers (M.A.P., J.A.M.) independently counted each slide using the same x,y coordinates, and the average of the two sets of counts was determined. The values obtained from two observers typically agreed with each other within 10%.

RESULTS AND DISCUSSION

Frequent and Often Independent Deletion of Distal and Proximal 8p Sequences

In an effort to define the 8p regions of deletion, we utilized PCR techniques to determine the allelic loss pattern of 26 highly polymorphic microsatellite sequences spanning 8p23-q12 for 85 paired normal and malignant tissues from cancerous human prostates. This analysis revealed loss of heterozygosity (LOH) of at least one 8p locus in 75% (64 of 85) of the tumors examined. This frequency is consistent with that obtained by other studies using more exacting or semiquantitative techniques, such as tissue laser microdissection (Vocke et al., 1996), phosphorimaging (MacGrogan et al., 1994), or fluorescence in situ hybridization (FISH; Macoska et al., 1994).

The deletion profiles of the 64 tumors with LOH are shown in Figure 1. Insufficient tissue precluded the complete analysis of six tumors (472, 129, 432, 476, 77, 484). Of the remaining 58 of 85 tumors, four (2, 454, 482, 504) demonstrated LOH for all 8p loci examined and were likely deleted for the entire 8p arm. An equivalent number of tumors (cases 23, 31, 322, and 488) were deleted for all 8p and 8q loci examined and were apparently reduced to monosomy for chromosome 8. Thus, eight tumors demonstrated substantial losses of chromosome 8 sequences. However, the majority of tumors examined in this study were deleted for extended regions rather than for all 8p sequences. Among the 64 cases with allelic loss, distal 8p sequences (8pter-p21), including D8S549, D8S1715, D8S1116, D8S1739, *NEFL*, D8S137, D8S1423, D8S540, D8S1125, D8S259, and D8S513 (indicated as "D" in Fig. 1), were deleted in 23 (27%) of tumors examined. Proximal 8p sequences (8p11-p12), including D8S1105, D8S535, D8S505, D8S87, D8S255, D8SD1118, D8S1121, and AFMA295zd5 (indicated as "P" in Fig. 1), were deleted in 16 cases (25%). Both distal and proximal sequences ("DP" in Fig. 1) were deleted contiguously in 25 cases (39%). Thus, separate or independent deletion of distal or proximal sequences (39 cases) was observed more frequently than contiguous deletion of both regions (25 cases) in these tumors. It should be noted that, in six cases (119, 127, 500, 486, 498, 129), the most distal portion of an otherwise contiguously deleted distal region was retained.

Identification of a Proximal 8p Breakpoint

A potential "breakpoint" may be identified from the data shown above in Figure 1. The boundary of this breakpoint appears somewhat "staggered" for many tumors (compare the proximal boundary for cases 498, 432, 224, and 242 with the distal boundary for cases 416, 434, and 460), roughly between D8S137 and D8S87. The *maximal extent* of this breakpoint can be defined as inclusive of loci D8S259, D8S513, D8S1105, D8S535, D8S505, and D8S87, which are all tightly linked and map within an approximately one megabase region between 64 cM and 65 cM on the genetic map (as reported by the Center for Genome Research at the Whitehead Institute for Biomedical Research). Although the

linkage order of these markers is uncertain, they are presented here in the order most consistent with the Whitehead Institute and with our deletion data.

To evaluate data from the current study in terms of what has been reported previously, it is critical to examine the definition of distal and proximal regions of 8p deletion in these studies. For example, several studies examined 8p loss using the marker D8S259, which comprises a part of the distal border of the 8p breakpoint defined in the present study. Observation of allelic loss patterns near and including D8S259 reported by MacGrogan et al. (1994) shows that 8 of 14 "group II" prostate tumors were deleted for 8p sequences immediately distal to or inclusive of D8S259, although the marker order D8S259-D8S137-D8S87 was used rather than the D8S137-D8S259-D8S87 order used here. Also, a few tumors were deleted separately for a region proximal to D8S137 in that study (MacGrogan, et al., 1994). Similarly, the study by Takle and Knowles (1996) demonstrated two 8p regions of deletion in a series of bladder carcinomas, one distal to and another proximal to loci inclusive of D8S259, although the marker order D8S505-D8S259-D8S513 was used rather than the D8S259-D8S513-D8S505 order used here. Inspection of their data also suggests the presence of a staggered "breakpoint" overlapping the proximal region of deletion and inclusive of loci D8S535, D8S505, D8S259, and D8S513. Moreover, realignment of these markers to conform with the order used in the current study does not essentially change the interpreation of their results.

A study by Trapman et al. (1994) demonstrated a large region of contiguous distal 8p allelic loss suggestive of a breakpoint proximal to nearby D8S137 (which maps distal to D8S259) in 9 of 26 prostates with 8p loss. Similarly, Farrington et al. (1996) defined one region of 8p loss distal to and another proximal to D8S137 in a series of colorectal carcinomas. A previous study from our laboratory also defined two separate regions of deletion distal to D8S137 as well as a third region of deletion proximal to that marker (Macoska et al., 1995). Vocke et al. (1996) described a major region of 8p deletion distal to and inclusive of D8S137, although it was more limited in extent than the major distal regions of deletion described in the other studies cited above.

Thus, it appears that all of these studies basically agree with a finding of one or more distal regions of 8p loss extending proximal to D8S137/D8S259, although they disagree on the presence of a separate region of 8p loss proximal to these markers. The basis of these discrepancies may be that 1) these studies did not examine the allelic loss pattern for markers mapping between D8S137/D8S259 and D8S87; 2) the physical order of markers mapping between D8S137/D8S259 and D8S87 is ambiguous, making data interpretation difficult; 3) the number of tumors examined in some studies may have been insufficient for the detection of discrete regions of allelic loss; and 4) there were probably differences in DNA preparation and gel evaluation, because fractional allelic loss frequencies range from 22% to 89% in these studies (MacGrogan et al., 1994; Trapman et al., 1994; Kagan et al., 1995; Macoska et al., 1995; Takle and Knowles, 1996; Farrington et al., 1996; Vocke et al., 1996).

It is interesting that the putative proximal 8p breakpoint identified here appears to be "staggered," in that it spans at least one megabase. Such "staggered" breakpoints have been observed for other chromosomes, notably the X chromosome, and have been implicated in the formation of dicentric isochromosomes (Wolff et al., 1996). Loss of 8p sequences apparently concurrent with 8q gain has been observed in prostate tumors by using FISH (Macoska et al., 1994) and comparative genomic hybridization (Visakorpi et al., 1995) techniques, potentially resulting from iso(8q) chromosome formation (Jenkins et al., 1997). The deletion data presented here are consistent with these scenarios.

Identification of a Homozygous Deletion at the D8S87 Locus

One tumor from case 2 (see Fig. 1) failed repeatedly to amplify at the D8S87 locus. To determine whether this was due to homozygous deletion at D8S87, we assayed paired normal and tumor tissue from case 2 by using multiplex PCR with primers to polymorphic sequences at D8S87 and primers to polymorphic sequences mapping distal (D8S505, D8S535) or proximal (D8S255, D8S1121) to D8S87. In all cases, DNA from case 2 tumor tissue failed to amplify at the D8S87 locus. Multiplex reactions showing coamplification of sequences at D8S87 and D8S1121 are shown in Figure 2 and show that two alleles that are 151 and 149 base pairs (bp) in length, respectively, amplify from the D8S87 locus in normal tissues from cases 2 and 23. However, both alleles fail to amplify in tumor tissue from case 2, and only the 149-bp allele amplifies appreciably in tumor tissue from case 23, suggestive of homozygous deletion (case 2) and heterozygous deletion (case 23) of D8S87 sequences, respectively. Coamplification of trinucleo-

tide repeat sequences at the D8S1121 locus reveals equivalent amplification of alleles 121 bp and 118 bp in size in normal tissue from case 2 and of alleles 124 bp and 115 bp in size in normal tissue from case 23. However, the corresponding tumors demonstrate heterozygous loss involving allele 121 in tumor 2 and heterozygous loss involving allele 124 in tumor 23. Furthermore, PCR analysis of loci producing amplicons larger than that of the D8S87 locus (e.g., the D8S549, D8S513, D8S505, D8S535, and D8S538 loci) was successful (see Fig. 1), suggesting that failure to amplify sequences from the tumor from case 2 DNA at the D8S87 locus was not due to DNA degradation. The ability to amplify sequences from one chromosome 8 homologue at the D8S1121 locus and the concurrent inability to amplify sequences from either chromosome 8 homologue at the D8S87 locus are consistent with the presence of a small homozygous deletion including the D8S87 locus in the tumor from case 2. This is the first report of homozygous deletion in this region of 8p in human tumors. Interestingly, the tumor from case 2 did not demonstrate homozygous loss at the D8S505 locus, which colocalizes to a 1.4 Mb CEPH yeast artifical chromosome, 898-G-11, suggesting that the region of homozygous loss may be less than 1.4 Mb in length.

To further verify the putative homozygous deletion at D8S87 in tumor 2, we performed FISH experiments. A human BAC clone, 303f13, containing sequences amplifiable at the D8S87 locus was labeled via nick translation with fluorescein-labeled dUTP (Amersham) and cohybridized with a rhodamine-labeled chromosome 8 centeromere-specific probe (Spectrum CEP8; Vysis) to the same normal and tumor tissues from case 2 (with the putative homozygous deletion at D8S87 by PCR analysis) and case 512 (with no deletion at D8S87 by PCR analysis). Two hundred nuclei from each of the four hybridized tissues were then enumerated for the number of D8S87-specific ("8p") and centromere-specific ("8c") probe signals (Table 1). Five types of nuclei were scored: those with 8p,8c counts equal to 1,1 (presumed monosomic for chromosome 8), 0,2 (presumed homozygous loss of 8p sequences), 1,2 (presumed heterozygous loss of 8p sequences), 2,2 (presumed disomic, with retention of 8p and 8c sequences), and other. Our data demonstrated a loss of 83% of D8S87-specific signal in the case 2 tumor compared with 25% in the corresponding normal tissue section. In contrast, case 512 demonstrated a loss of 38% of D8S87-specific signal in both normal and tumor tissue sections (Table 1). These data are consistent

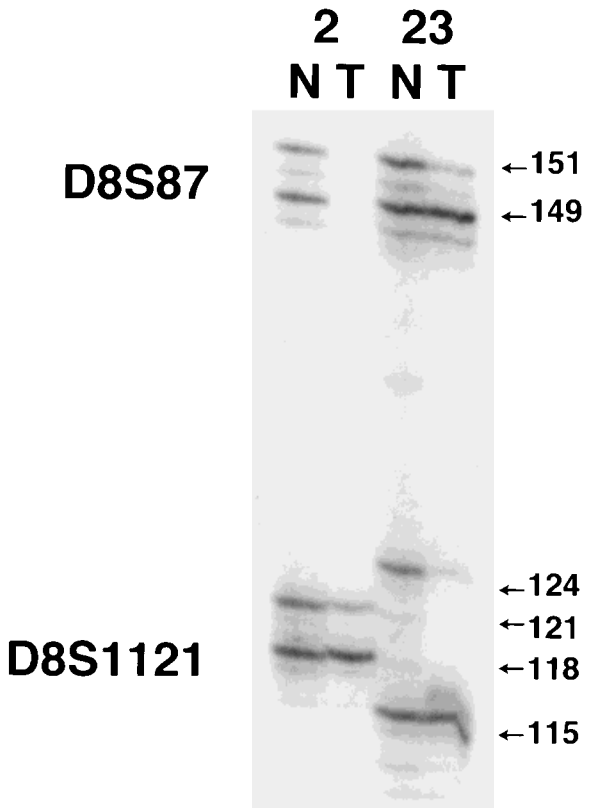

Figure 2. Homozgyous deletion at the D8S87 locus in a prostate tumor. Autroradiograph of a gel showing the results of multiplex polymerase chain reaction analysis using primers to the D8S87 and D8S1121 loci in paired normal (N) and tumor (T) tissues from cases 2 and 23. Alleles that are 151 and 149 base pairs (bp) in length, respectively, from the D8S87 locus amplify equivalently in normal tissues from cases 2 and 23. However, both alleles fail to amplify in tumor tissue from case 2, and only the 149-bp allele amplifies appreciably in tumor tissue from case 23, suggesting a homozygous deletion (case 2) and a heterozygous deletion (case 23) of D8S87 sequences, respectively. Coamplification of trinucleotide repeat sequences at the D8S1121 locus reveals equivalent amplification of 121-bp and 118-bp alleles in normal tissue from case 2 and of 124-bp and 115-bp alleles in normal tissue from case 23. However, the corresponding tumors demonstrate heterozygous losses involving a 121-bp allele in the tumor from case 2 and a 124-bp allele in the tumor from case 23. The ability to amplify sequences from one chromosome 8 homologue at the D8S1121 locus and the concurrent inability to amplify sequences from either chromosome 8 homologue at the D8S87 locus are consistent with the presence of a small homozygous deletion, including the D8S87 locus in the tumor from case 2.

with the PCR data, suggesting complete or nearly complete loss of D8S87-specific sequences in case 2, with retention of the same sequences in case 512. The degree of apparent loss of D8S87-specific sequences in the normal tissues of case 2 (20%) and in both tissues from case 512 (33–36%) is consistent with that obtained from sectioned tissue and is likely to be due to "slice artifact," as reported previously (Wolman et al., 1993; .Macoska et al., 1994; Jenkins et al., 1997).

Homozygous deletion has been interpreted in other studies as indicating the physical localization

TABLE 2. Correlation of 8p Deletional Domains With Clinical and Pathological Parameters

Parameter	Type			
	Distal	Distal + proximal	Proximal	None
Stage				
LN+ (n = 11)	3	3	1	4
T4 (n = 6)	3	1	1	1
T3 (n = 32)	9	8	6	9
T2 (n = 36)	8	13	8	7
Grade				
5,6 (n = 20)	6	5	5	4
7 (n = 38)	9	12	8	9
8,9 (n = 27)	8	3	3	8
Preoperative PSA[a]				
0.0–4.0 (n = 12)	1	4	5	2
4.1–10.0 (n = 28)	11	5	3	9
10.1–20.0 (n = 19)	3	7	4	5
>20.1 (n = 26)	8	9	4	5
Age (years)				
39–54 (n = 15)	4	3	2	6
55–65 (n = 38)	9	15	7	7
66–75 (n = 31)	9	7	7	8
ND (n = 1)	1			

LN+-metastatic to regional lymph nodes
[a]PSA, prostate-specific antigen; ND, no date.

of tumor-suppressor genes, and, most recently, it aided in the molecular cloning of two such genes, *DPC*-4 (Hahn et al., 1996) and *PTEN* (Li et al., 1997). Therefore, evidence of homozygous loss of the D8S87 locus in a human prostate tumor is consistent with the hypothesis that a proximal 8p tumor-suppressor gene is important in prostate tumorigenesis and maps this tumor-suppressor gene more precisely to a location at or near the D8S87 locus.

Correlation of 8p Deletional Regions with Clinical and Pathological Parameters

The frequency of distal, distal+proximal, proximal, and lack of allelic loss was correlated with tumor pathologic stage, combined Gleason score (grade), preoperative serum PSA values, and age at diagnosis of the PCa patients in this study group. A breakdown of the number of tumors in each category is provided in Table 2. There were no statistically significant differences between the number of tumors in each of the four categories (distal, distal + proximal, proximal, and lack of allelic loss) in relation to pathologic stage ($P = 0.924$), grade ($P = 0.890$), preoperative PSA ($P = 0.224$), or age at diagnosis ($P = 0.558$). Because ethnic origin was not recorded for 28 of 85 (33%) patients included in this study, statistical evaluation of that variable was not attempted.

CONCLUSIONS

In conclusion, this study identifies the physical location of a proximal 8p breakpoint, and it demonstrates a novel homozygous deletion of the proximal D8S87 locus in human prostate tumors. The data are consistent with the hypothesis that a proximal 8p tumor-suppressor gene that is important for prostate tumorigenesis maps at or near the D8S87 locus and should facilitate the isolation and cloning of candidate tumor-suppressor genes from this region.

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