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Advances in Brief

Prognostic Impacts of Cytogenetic Findings in Clear Cell Renal Cell Carcinoma: Gain of 5q31–qter Predicts a Distinct Clinical Phenotype with Favorable Prognosis

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Abstract

To evaluate the prognostic significance of cytogenetic findings in clear cell renal cell carcinoma (RCC), cytogenetic results of 118 primary RCCs were evaluated in relation to classical indicators of prognosis and overall survival. Losses in 3p (98.3%) were most prevalent and included 32 (27.6%) monosomies of chromosome 3 and 84 (72.4%) structural aberrations involving 3p, of which 36 were unbalanced translocations, der(3)t(3;5)(p11–p22;q13–q31), resulting in duplication of 5q sequences. Patients with gain of 5q31–qter resulting from either polysomies or structural rearrangements of 5q, the most frequent of which was der(3)t(3;5), had a significantly better outcome than those without this aberration ($P = 0.001$). There was no association between gain of 5q or der(3)t(3;5) and any of the well-known variables for prognosis, including low *versus* high clinical stage and grade of malignancy. Among additional chromosomal aberrations, loss of chromosome 9/9p was associated with distant metastasis at diagnosis ($P = 0.006$). The data indicate that gain of 5q identifies a clinically favorable cytogenetic variant of clear cell RCC and demonstrate the impact of specific chromosome aberrations as additional prognostic indicators in clear cell RCC.

Introduction

The classification of renal epithelial tumors has steadily evolved since the initial recognition of them as "hypernephromas." Emerging from former histomorphologically oriented distinctions, recent classifications have been advanced on the basis of histo- and cytomorphological aspects, as well as histogenetic and cytogenetic aspects, distinguishing between clear cell RCC[2] and papillary RCC both derived from the proximal tubule, chromophobic RCC and oncocytoma from the cortical collecting duct, and Bellini duct carcinoma from the medullary collecting duct (1–3). Clear cell RCC is the most common histological type constituting approximately 70–80% of all epithelial neoplasms of the kidney. Genetically, the clear cell type is distinct from other categories in that most RCCs with clear cell cytomorphology are characterized by loss of the short arm of chromosome 3 (4, 5). However, within this seemingly homogeneous category, tumors show varying clinical behavior. Fewer than 50% of the patients are cured by surgical treatment; the remaining patients either have a metastatic disease at the time of diagnosis or develop metastasis within 5 years after surgery, with poor survival prospects. At present, prediction of likely tumor behavior is best determined by the clinical stage and nuclear grade at the time of surgery (6, 7). Further markers for prognosis are increasingly being suggested by cytogenetic and molecular techniques, indicating that the frequency of deletions at chromosome arms 8p, 9p, and 14q is associated with higher stage and grade of clear cell RCCs (8, 9), and loss of 9p is correlated with recurrence (10). Another study suggests that the degree of cytogenetic complexity, rather than the mere presence of specific aberrations, is a superior predictor of likely outcome, in that patients with few karyotypic changes had a prolonged disease-free survival compared with those with more than five changes (11). These findings imply that karyotypic complexity and recurrent cytogenetic aberrations may be of prognostic significance. On the other hand, considerable clinical heterogeneity exists within the clear cell RCC category, and it is quite possible that the current histomorphological classification lumps together genetically distinct subgroups with different clinical phenotypes. Genetic heterogeneity within the clear cell RCC category is already evident in the variable presence of chromosomal translocations, in particular der(3)t(3;5), deletions, and numerical abnormalities involving chromosome 3. However, the impact of chromosome aberrations as a possible basis for a clinically useful subdivision has thus far never been evaluated properly. Here we examined a consecutive series of 118 patients with primary clear cell RCC to investigate whether cytogenetic changes can add valuable prognostic information.

Materials and Methods

Patients and Tumor Samples. The series originally comprised 143 adult patients with clear cell RCCs treated at the Departments of Urology at the University Hospital of Aachen, Germany and at the Georg August University Hospital of Göttingen, Germany during the period from March 1989 to June 2000. Twenty-five patients whose tumors could not be karyotyped because of failure to grow in cell culture (18 cases) or too few analyzable metaphases (7 cases) were excluded from the study. For the remaining 118 patients, follow-up data were obtained through files from the respective clinics, local tumor centers, and general practitioners of the patients. These data were reviewed to determine whether the death of a patient was tumor related or the result of other causes. About 10–20 H&E-stained tumor sections were evaluated to establish the diagnosis of clear cell RCC and to determine the growth pattern, grade of malignancy, and the presence of infiltrative growth into fat, fascia, or veins. Tumors were staged according to the TNM system (12). For grade of malignancy, a three-grade scale system was used.

Cytogenetic Analysis. For classical cytogenetic analysis, viable and cellular tumor samples were excised immediately after surgery by experienced pathologists; one part was snap frozen and stored at −80°C, and the other part was used for short-term culture and chromosome analysis. Chromosomes were banded using routine G-banding and 4′,6-diamidino-2-phenylindole-banding techniques. For cases investigated after 1996, image acquisition and analysis of

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[2] The abbreviations used are: RCC, renal cell carcinoma; CI, confidence interval; TNM, Tumor-Node-Metastasis.

Table 1 *Clinicopathological and cytogenetic findings of 118 primary clear cell RCCs*

No.	Age/Sex[a]	Size	Stage	Grade	Follow-up in months	Karyotype
1	74/F	8	III	2	NA	44-46,XX,der(3)t(3;5)(p12;q15),-14,del(16)(q22),+21[cp30]/86-92,idemx2[cp3]
2	72/M	5	I	1	Alive, 12	41-45,XY,-3,der(6)t(3;6)(q21-23;q21)[cp18]/84-90,idemx2[cp8]/41-45,idem,-15[cp4]
3	50/M	4.5	I	1	NA	39-45,XY,-3[cp8]/46-49,XY,+5,+10,+12[cp6]/39-46,XY[cp38]
4	52/M	5	I	2	Alive, 9	44-45,X,-Y,der(3)t(2;3)(q23;p13)[cp4]/44-45,idem,del(14)(q21q24)[cp24]
5	77/M	6	III	2	Alive, 12	71-77<4n>,XXYY,-1,-2,-2,-3,-3,+5,-6,der(7)t(3;7)(q21;q32)x2,-8,i(8)(q10)x2,-9,-9,-10, -11,-13,-14,-14,-15,-15,-17,i(17)(q10),-18,-18,-19,add(19)(q13),+21,+21, +mar1[cp6]/45-47,X,-Y[6], +7[9][cp11]/47,XY,+X[2]/46,XY[9]
6	63/M	16	III	2	Alive, 8	75-81<4n>,XXY,-Y,-1,der(3)t(3;5)(p13;q13)x2,-6,-6,-8,-9,del(10)(q23)x2,-11,-13,-14, -18,-20[cp9]/73-85,idem,-14[cp19]
7	58/M	8	II	2	Alive, 11	41-45,Y,del(X)(p22),der(3)t(3;8)(p12;q11),-6,-8,add(15)(q2?6)[cp10]/45,idem, +der(3)t(3;8)(p12;q11)[1]/39-44,idem,-Y,+der(3)t(3;8)(p12;q11),del(9)(q13)[cp8]/ 40-45,idem,-Y,+der(3)t(3;8)(p12;q11),+5,del(9)(q13)[cp6]/43-47,-Y,+der(3)t(3;8) (p12;q11),+5,del(9)(q13),+17[cp2]/39-43,idem,-Y,+5,del(9)(q13)[cp21]/46,XY[2]
8	79/F	6	III	2	Alive, 2	44-45,XX,dic(3;15)(p12;q12),ins(5)(q1?4),-15[cp5]/88-90,idemx2[cp2]/42-45,idem, del(4)(q22q28)[cp7]/90,idemx2,del(4)(q22q28)x2[2]/43-44,idem,-8,der(22)t(8;22) (q11;p13)[cp13]/42-46,idem,-8,del(14)(q22),-16,+der(22)t(8;22)(q11;p13), +der(22)t(8;22)(q11;p13)[cp17]/42-44,idem,+dic(3;15)(p12;q12)[cp2]/46-48,XX[cp2]
9	67/F	6.5	I	2	Alive, 15	42-47,XX,add(1)(p36),add(2)(q37),del(3)(p12),-14,-22[cp15]/44,idem,add(7)(q36) [cp6]/41-44,idem,add(6)(p25),add(7)(q36)[cp2]/44-45,idem,+7,add(7)(q36), add(12)(q24)[cp2]/45-46,idem,add(X)(q27),+12,+20[cp2]/46-49,idem,+12,add(16) (p13),+20,+22[cp2]/89-92,idemx2,+12,+12,der(17)t(3;17)(q11;p13)x2,+20,+20[cp2]
10	71/M	9	IV	3	NA	74-82,XX,-Y,-Y,-1,-3,-3,der(6)t(3;6)(q11;q11)x2,-8,-9,-10,-14,-18[cp15]
11	59/F	10	IV	3	DOTD, 4	45-46,XX,del(3)(p12),der(6)t(6;13)(q12;q12),-13,+20[cp2]/41-45,idem,-10[cp16]
12	43/M	12	II	2	Alive, 4	70-80,XXYY,der(2)t(2;8)(q35;q13)x2,der(3)t(3;5)(p21;q22)x2,-5,del(6)(q23),-8,-9,-14, -15,-17,-21[cp6]/161,idemx2[1]/73-82,idem,-5,der(11)t(8;11)(q13;p13)x2,-14,-15, add(17)(p11),-18,+mar1,+mar1[cp19]
13	47/M	13	IV	2	Alive, 3	40-43,XY,der(3)t(3;5)(p13;q1?5),add(4)(q3?),der(6)t(6;8)(p12;q12),-8,-9,der(10)t(6;10) (p12;q26),-14[cp7]/45,XY,-20[1]
14	56/M	6	III	2	Alive, 3	43-44,XY,-3,der(8)t(8;14)(q13;q13),-14,der(18)t(8;18)(q13;q23)[cp4]/43-46,XY[cp9]
15	72/M	8	III	2	Alive (DP), 7	40-47,X,-Y,+2,der(3)t(3;13)(p14?;q13),+5,+7,-13,t(13;21)(p11;q21)[cp17]/45,X,-Y, dup(13)(q12q14)[1]
16	70/F	3.5	I	1	NA	37-46,XX,der(3)t(2;3)(q2?4;p13)[cp17]/46,XX[4]
17	68/M	9	III	2	Alive, 1	45-46,XY,ins(3)(p22p24)[cp8]/43-46,X,-Y[cp12]/42-47,XY[cp20]
18	76/M	15	IV	3	Alive, 3	43-48,XY,der(3)t(3;5)(p13;q22),-6,+7,-8,add(11)(q25),+12,-14,+mar1[cp6]/48-49,idem, +der(3)t(3;5)(p13;q22),-6,+7[cp3]/45-46,XY[cp3]
19	62/M	5	I	1	Alive, 1	41-44,XY,der(3)t(3;5)(p13;q15),-14[cp4]/82-90,idemx2[cp3]
20	79/M	6	I	2	Alive, 4	39-46,XY,der(3)t(3;5)(p22;q14)[cp7]/46,idem,der(X)del(X)(p21)ins(X)(q11)[cp11]/ 45,X,-Y[cp14]/43-46,XY[cp23]/89-92,XXYY[cp3]
21	52/M	6	I	2	DOO, 109	43-45,X,-Y,del(3)(p14~21),+5[cp28]
22	63/F	9	III	2	Alive, 70	43-47,XX,der(1)t(1;5)(p31;q21),+der(1)t(1;5)(p31;q21),+2,del(3)(p13~21),+7,-8,-14, -18[cp12]/45,idem,-19[3]
23	64/F	8	II	1	NA	42-44,XX,-3,der(4)t(3;4)(q13;p15),-6,-14[cp72]/42-44,idem,i(8)(q10)[cp15]/ 43,idem,del(8)(p11)[5]/42,idem,-8[cp3]
24	62/F	8.5	IV	2	DOTD (DP), 41	45,XX,der(1)t(1;3)(q12;q21),der(2)t(1;2)(q12;q37),-3[cp6]
25	54/F	6	I	1	Alive, 125	43-44,XX,-3,der(4)t(3;4)(q13;p15),-14[cp12]
26	63/F	3.5	I	1	Alive, 120	43-50,X,-X,der(3)t(3;8)(p12;q11),+5,+7,-8,+11,+16,+18[cp10]
27	68/M	7	I	1	Alive, 124	42-45,XY,add(1)(p36),del(2)(p13),der(3)t(3;8)(p11;q11),-6,-8,+mar1[cp21]
28	68/M	5.5	I	1	DOO, 71	46,XX,der(3)t(3;7)(p11;q11),add(4)(p15),add(9)(q34)[cp50]/44-46,idem,-17[3], -19[3][cp22]
29	71/F	3.5	I	1	Alive, 41	44-47,XX,add(3)(p13)[cp47]/45-46,idem,add(19)(p13)[cp4]/45-47,idem,del(14)(q22) [cp14]/46-47,idem,+5,del(14(q22)[cp14]/45-46,idem,add(2)(p25),del(14)(q22)[cp9]/ 46,XX[6]
30	55/F	3	I	2	NA	42-45,XX,der(1)t(1;5)(p34~36;q15),del(3)(p13),+7,-9,-14,-15,+r[cp13]/44-45,idem,-8, +mar1[cp2]/45-46,idem,+der(1)t(1;5)(p34~36;q15)[cp7]/48-49,idem,+der(1)t(1;5) (p34~36;q15),+2,+17[cp3]/72-75,idemx2,-X,-4,-5,-5,-6,-7,-8,-10,-11,-13,-13,-16,-22,

[a] F, female; M, male; NA, not available; DOO, died of other causes; DP, disease progression; DOTD, died of tumor disease.

metaphase spreads were performed on a Vysis Quips Genetics Workstation using the Quips Karyotyping Software (Vysis, Bergisch-Gladbach, Germany). Clonality criteria and karyotype descriptions follow the recommendations of ISCN (13). For practical purposes, cytogenetic abnormalities were expressed as net changes of chromosomes or chromosome arms in relation to even ploidy levels. By convention ploidy levels were classified as pseudodiploid (modal number, 46), hypodiploid (modal number, <46), hyperdiploid (modal number, 47–57), and polyploid (modal number, ≥58).

Statistical Analysis. The associations between clinicopathological variables (tumor size, TNM stage, clinical stage, and grade of malignancy) and cytogenetic abnormalities were evaluated using the two-sample Wilcoxon test and Fisher's exact test for contingency tables. Survival rates were plotted using the Kaplan-Meier method. Clinicopathological variables and the most common cytogenetic abnormalities were studied for their association with overall survival and tested with the Mantel-Haenszel test (log-rank test) for censored data.

Table 1 *Continued*

						-r,-r[cp2]/75-76,idemx2,-X,-4,-5,-5,-6,-7,-8,-10,-11,-13,-13,-22,-r,-r[cp2]/74,idemx2, -X,-2,-4,-5,-5,-6,-7,-8,-10,-11,-13,-13,-22,-r,-r[cp2]/73-74,idemx2,-X,-2,-4,-4,-5,-5,-6, -7,-8,-10,-11,-13,-13,-22,-r,-r[cp7]/70-74,idemx2,-X,-2,-4,-4,-5,-5,-6,-7,-7,-8,-10,-11, -13,-13,-22,-r,-r[cp5]
31	75/M	6	I	3	NA	46-47,XY,del(3)(p13),+5,-14,+16[cp3]
32	52/F	3.5	I	1	Alive, 119	45,XX,der(3)t(3;5)(p12~13;q22~23),-14[2][cp3]
33	38/F	4.5	I	1	Alive, 117	45,X,-X,der(3)t(3;5)(p12;q31)[13]/106,idemx2[1]/46,idem,+2[1]/44-53,idem,+X,+2, +der(3)t(3;5)(p12;q31),+8,+12,+16,+19,+20[cp8]
34	80/F	4	I	2	DOO, 100	42-45,XX,der(2)t(2;5)(q32;q21),-3,t(3;16)(q12;q22),+7,-9,-14,der(22)t(3;22) (q11;p13)[cp62]/40-47,idem,-5[3],-7[4],-11[3],-12[4],-17[4][cp15]
35	66/M	7	III	3	DOTD (DP), 8	43-48,XY,del(3)(p14)[cp72]/42-46,idem,add(1)(p34~36),del(11)(q14q22), add(13)(p11)[cp3]
36	69/M	11	II	2	Alive (DP), 112	46,XY,der(3)t(3;5)(p12~14;q15~22)[cp2]
37	72/F	2.5	I	2	DOO, 86	45,X,-X,add(3)(p21)[11]/43-45,idem,-18[3][cp12]
38	71/M	6	IV	3	DOTD, 3	43-45,X,-Y,add(3)(p12)[cp8]
39	78/F	6	I	2	Alive, 106	42-45,XX,-3,der(6)t(5;6)(q15;q21),add(12)(p13),-14,der(20)t(3;20)(q13;p12),-21, +mar1[cp43]/44,idem,add(9)(q34)[2]/43-44,idem,der(9)t(1;9)(q12;q12)[cp7]/ 86,idemx2,-der(6)t(5;6)(q15;q21),-9,-16,-16,-22,-22,-mar1,+3mar[1]
40	56/M	8	II	1	DOTD (DP), 15	43-45,X,-Y,del(1)(p22),+del(1)(p22),del(3)(p14),-8,-14,add(15)(p13)[cp29]/ 42,idem,-del(1)(p22),-4[4]/42,idem,-del(1)(p22),-4,-10,+mar1[8]
41	69/M	4	I	2	DOO, 56	46,X,-Y,-3,+5,inv(6)(p12q22),+7,del(9)(q34),-20,+mar1[11]/42-46,idem,-6,-10,-18, der(22)t(18;22)(q11;q11),+mar2,+mar3[cp65]
42	63/F	5	IV	2	DOTD, 23	46,XX,del(3)(p14)[1]/35-46,idem,-14[cp12]/44-46,XX[cp3]
43	81/F	2.5	I	1	DOO, 6	42-43,X,-X,add(3)(p12),del(6)(q16),t(7;16)(q11;q11),-9,-14[cp9]
44	67/M	10	IV	1	DOTD (DP), 5	43-47,XY,add(3)(p13)[cp80]/92,idemx2[cp5]/43-46,idem,add(19)(q13)[cp6]/ 92,idemx2,add(19)(q13)x2[1]/47,idem,+13[2]/42-46,XY[cp2]
45	71/F	9	III	2	DOO, 20	44-47,XX,del(3)(p14)[cp83]/92,idemx2[cp2]/44-45,idem,-14[cp5]
46	68/M	3	I	1	DOO, 83	44-45,X,-Y,+5,-14,-18[cp8]/44-45,idem,+20[cp10]
47	64/F	6	I	2	Alive, 95	43-47,XX,der(3)t(3;5)(p13;q22),del(8)(p21),-14[cp12]
48	49/F	6	I	1	Alive, 100	45-47,XX,-3,+mar1[cp19]
49	38/M	4.7	I		NA	64-72<4n>,XXYY,der(1)t(1;3)(p21;q13)x2,-2,-3,-3,-3,-4,-6,-6,-8,-8,-9,-9,-10,-10,-11, del(12)(q24)x2,-13,-14,-14,-15,-17,-18,-18,-21,-22[cp16]/136-138,idemx2[cp2]/ 47,XY,+Y[2]/46-47,XY[cp2]
50	44/M	2.5	III	1	Alive, 78	74<4n>,XXY,-Y,-1,-2,-3,-3,-4,+5,ins(5)(p12q13)x3,-6,-6,-10,-10,-11,-11,-13,-14,-14, -15,-16,-18,-19,-19,+mar[1]/43-46,XY[cp25]
51	32/F	9	II	1	Alive, 85	43-47,XX,der(3)t(3;5)(p14;q13)[cp15]/92,idemx2[1]/42-48,idem,-21[cp3]/ 42-46,idem,-14[cp8]/40-44,idem,-14,-22[cp3]
52	62/M	3	I	1	Alive, 86	39-45,X,-Y,del(1)(p34),+2,add(3)(p13),-8,-14,+16[cp24]/43-47,idem,+5[cp6]/46,XY[4]
53	57/F	10	III	3	DOTD (DP), 62	45-46,XX,r(3)[cp9]/44-45,idem,-14[cp9]/45-48,idem,+del(1)(p13),+5,+7,-14[cp9]/ 48-49,idem,+del(1)(p13),+5,+7,i(8)(q10),-14[cp3]
54	66/F	5	III	1	Alive, 86	45-46,XX,+der(1)t(1;2)(p11;q11),der(3)del(3)(p14)t(1;3)(q25;q26), der(6)t(5;6)(q14;q21),-14[cp31]/92,idemx2[4]
55	78/F	7	I	1	Alive, 84	46,XX,der(3)t(3;5)(p21;q22)[cp21]/46,idem,del(14)(q24)[cp9]/46,XX[1]
56	67/M	4.5	I	2	Alive (DP), 83	41-44,XY,del(3)(p13),-6,del(8)(p21),+del(8)(p21),-9,-10,+12,-13,-14,-18,+mar1[cp32]
57	64/F	6	I	1	DOO, 78	43-47,XX,del(3)(p14)[cp36]
58	74/F		III	1	NA	45-46,XX,der(3)t(3;5)(p12;q23~31)[cp28]/42-45,idem,-14[cp9]
59	77/F	5	III	1	DOO, 45	43-47,XX,der(3)t(3;5)(p13;q15),-14,+mar[cp52]/43-45,idem,-8[cp4]/42-44,idem,-9[cp3]
60	72/M	4	I	1	Alive, 95	43-45,XY,-3,der(4)t(3;4)(q21;q28)[cp30]/84-93,idemx2[cp4]/85-88,idemx2,-8,-8[cp3]/ 84-88,idemx2,-8,-8,-9[cp13]/86-87,idemx2,-8,-8,-9,-9[cp2]/82-85,idemx2,-8,-8,-9,-14, -14[cp3]/82-85,idemx2,-8,-8,-9,-13,-13[cp2]
61	63/F	3	I	1	Alive, 83	44-47,XX,r(3)[cp56]/45-46,idem,del(8)(p11.2)[cp3]/46,idem,t(7;20)(q22;q11.2), del(14)(q24)[cp2]
62	70/M	5.5	III	1	Alive (DP), 81	50-53,X,-Y,der(3)t(3;5)(p12;q23),+der(3)t(3;5)(p12;q23),+5,+7,+11,+12,+16,+16, +20[cp19]/53-54,idem,+8[cp5]
63	62/M	2.5	I	1	Alive, 72	43-44,XY,der(3)t(3;5)(p14;q31),-6,-9[cp23]
64	59/F	7.5	II	1	Alive, 92	45,XX,del(2)(q23),der(3)t(2;3)(q23;q29),der(3)t(2;3)(q23;q29)t(3;5)(p14;q23),-14, add(17)(q25)[4]/46,idem,+22[13]
65	62/F	8	III	1	Alive, 72	52,XX,+X,+2,der(3)t(3;14?)(p14;q13?),+5,+12,+20,+21[3]
66	64/F	10	II	1	Alive, 80	44-46,XX,del(3)(p14)[cp29]

Results

Patients and Follow-Up. Of the 118 patients whose tumors could be karyotyped, there were 60 men and 58 women with a mean age at diagnosis of 63.5 years (median, 64 years; range, 32–81 years). There was no family history of RCC; also, all patients were *de novo* RCC cases that had no history of preexisting renal tumors. All patients were treated surgically, either by total nephrectomy or partial resection, and no one had received chemotherapy or radiation therapy prior to surgery. Follow-up data were available for 104 patients with a mean follow-up duration of 42 months (median, 26 months; range, 1–125 months). Of these 104

Table 1 *Continued*

67	61/M	6	I	2	DOTD (DP), 71	64-65<4n>,XX,der(X)t(X;13)(p11;q12)x2,-1,-1,-2,-3,-3,-3,-5,-6,-6,-8,-8,-9,-10,-11,-12,-13,-14,-14,-15,-15,-16,-17,-17,-18,-19,-20,-21,-22[cp4]/127,idemx2,-21,-21,+mar[1]/59-64,idem,-10,-22[cp6]/53-55,idem,-2,-4,-11,-13,-18,-19,-22[cp4]/46,XX[cp8]
68	70/F	4.5	I	1	Alive, 86	41-47,XX,-3,der(3)t(3;4)(q13;q31)[cp43]/88-90,idemx2[cp4]/44-47,XX,+Y[2][cp3]
69	66/F	16	III	3	Alive, 78	42-47,XX,der(1)add(1)(p36)del(1)(q41),der(3)t(3;5)(p12;q31)[cp3]/46,idem,+7,-14[4]/43-45,X,-X[cp17]/47,XX,+20[2]/46,XX[3]/45-48,XX[cp3]
70	69/F	11	II	2	Alive, 78	42-45,X,t(X;12)(p22;q13),-3,add(22)(p11)[cp6]/38-44,idem,-14[cp7]/45-48,XX[cp18]
71	65/M	7	I	2	DOO, 42	44-47,XY,del(3)(p13),del(8)(p11),-9,+12,-13,-14,+mar1,+mar2[cp29]
72	60/M	4	IV	2	DOTD, 38	44-45,XY,add(3)(p11),-14[cp5]/86-88,idemx2[cp2]/41-45,idem,-9[cp17]/46,XY[1]
73	62/M	9	III	2	Alive, 71	72-76<4n>,XXYY,-1,der(3)t(3;5)(p12;q21~22)x2,-6,-8,-8,-9,-9,-10,-13,-13,-14,-14,-15,-17,-18,-18,-22[cp6]/72-78,idem,+mar1[cp4]
74	68/F	3	I	1	Alive, 67	41-44,XX,add(3)(p13),-6,-8[cp33]
75	64/F		IV	2	DOTD (DP), 13	43,XX,der(3)t(3;6)(p11;p11),-6,-9,-13[cp2]/41-45,idem,+1[cp6]/41-42,idem,-22[cp3]/45-46,XX[cp2]
76	57/M	9	IV	2	DOTD (DP), 28	42-45,X,-Y,del(3)(p12),der(6)t(5;6)(q13;q27)[cp5]/40-43,idem,add(1)(p13),-8,der(13)t(13;14)(q21;q11),-14[cp10]/42-43,idem,add(1)(p13),del(4)(q21),-8,der(13)t(13;14)(q21;q11),-14[cp3]/40-44,idem,der(1)t(1;14)(p32;q24),del(8)(p21),del(12)(p11),-14[cp6]
77	75/M	6	III	3	DOTD (DP), 12	60-67<4n>,der(X)t(X;2)(q28;q21)x2,-Y,-Y,der(1)t(1;8)(p12;q12),-2,-3,-4,-5,-6,-6,-8,-8,-9,add(9)(p11),-10,-11,add(12)(q12),-13,-14,-15,-16,-16,-17,-17,-18,-18,-19,-19,-21,-21,-22,-22,-22,+mar1,+mar2,+mar3[cp30]
78	58/F	3	I	2	Alive, 42	38-45,X,-X,der(3)t(3;5)(p12;q15)[cp7]/90,idemx2[1]/84-88,idemx2,-18,-18[cp2]/43-44,idem,-14,-18,+mar[cp8]
79	59/F	9	IV	2	DOTD (DP), 24	44,XX,del(1)(p31p31),-3,-9,-14,i(17)(q10),+mar1[cp2]/42-44,idem,-4[cp32]/87,idemx2,-4,-4,+mar[1]
80	62/F	4.5	IV	2	DOTD, 31	36-44,X,-X,add(3)(p21),+7,-9,-14,-18[cp4]/81,idemx2,+mar[1]/44-45,idem,+ins(8)(q21)[cp3]/91,idemx2,+ins(8)(q12)x2,i(12)(q10),+4mar[1]
81	69/F	6	III	2	Alive (DP), 31	42-45,XX,der(3)t(3;5)(p14;q15),-6,der(16)t(6;16)(p11;q11)[cp15]/88-92,idemx2[cp2]/43-44,idem,der(1)t(1;5)(p35;q15),del(4)(q22),-14[cp8]/46,XY[1]
82	61/M	4	III	2	DOTD (DP), 48	45-51,XY,-3,+5,+20,+22[cp4]/46,XY[1]
83	79/M	5	III	2	Alive, 39	46-52,X,-Y,+del(1)(p13),+2,der(3)t(3;5)(p11~13;q15~22),+5,del(11)(q21),+12,-14,+add(17)(p11),+22,+mar1[cp14]/51-52,idem,+del(11)(p11.2)[cp2]/46,XY[1]
84	38/M	3.5	I	2	Alive, 24	46-47,XY,-3,-14,+18,+21,+mar1[cp34]
85	47/F	4	III	1	Alive, 22	43-46,XX,der(3)t(3;5)(p14;q15)[cp23]/46,XX[2]
86	77/F	10	IV	2	DOTD (DP), 24	88,XXXX,del(1)(p13)x2,der(3)t(3;5)(p12;q13)x2,add(5)(p15)x2,ins(5)(q11)x2,-6,-6,add(7)(p13)x2,der(10)t(1;10)(q12;q26)x2,-13,-14,-14,-18,+mar1,+mar1[cp5]/84-89,idem,+2[7],-19[6],-mar1[3][cp12]
87	62/M	5	III	2	Alive, 26	43-45,X,-Y,der(3)t(3;5)(p12;q14),add(15)(p13)[cp23]
88	66/M	1.5	I	2	Alive, 33	41-46,XY,-3,der(4)t(4;5)(p16;q13),+7,-14,+mar1[cp8]/45-46,idem,-8[9],t(10;16)(q24;q24)[8],+mar2[cp9]/46,XY[7]/44-45,X,-Y[cp4]
89	67/F	8	IV	2	DOTD, 11	39-46,XX,der(3)t(3;8)(p11;q11),-8,+12[cp11]
90	69/M	6	I	2	Alive, 16	69-84,XXYY,del(1)(p22)x2,add(3)(p12~14)x2,-6,-6,-6,-8,-9,-10,-14,-14,-15,-17[cp2]/44-45,X,-Y[cp2]/44-46,XY[cp10]
91	49/M	4	IV	2	Alive, 30	40-45,X,-Y,der(3)t(3;7)(p13;q11.2)[cp11]/36-44,idem,-9[cp4]/84-88,idemx2,-9,-9[cp3]/80,idemx2,-10,-12,-12,-13,-14,-14,-17,-21,-21[1]
92	61/F	2.7	III	1	Alive, 32	40-47,XX,add(3)(p21),+5[cp20]/45-46,idem,-14[cp3]/45,XX,-17[1]
93	51/M	3.5	III	2	Alive, 21	44-45,XY,dup(2)(q?q?),der(3)t(3;5)(p11~13;q14~22),-14[cp4]/82-89,idemx2[cp5]/45-46,XY[cp2]
94	56/F	5	IV	2	DOTD (DP), 8	43-46,XX,-3,der(6)t(3;6)(q13;q16),+7[cp22]/45-46,idem,-14[cp3]/46,XX[3]/92,XXXX[1]
95	81/F	4	I	2	DOTD (DP), 26	40-45,XX,del(3)(p12),i(8)(q10),-9,-14,+16[cp7]/40-47,idem,+5[cp9]/83,idemx2,-X,-X,-del(3)(p12),-5,-5,-9,-11,-12,-20,-20,+mar[1]
96	53/F	8	IV	3	DOTD, 12	51,XX,t(2;7)(p13;p22),del(3)(p12),+5,+7,i(8)(q10),add(9)(p23),+12,del(17)(p11),+del(17)(p11),+20[1]/49-50,idem,der(14)t(5;14)(q11;p13),-15[cp2]/41-46,XX[cp9]
97	76/M	3	I	1	NA	69-76<4n>,XX,-Y,-Y,-1,-3,-3,-4,+5,+5,-6,add(7)(p24)x2,-8,-8,-9,add(9)(p13),-10,-13,-14,-15,-16,-18,-22,-22[cp10]/73-77,idem,+3,+8,-14[cp2]
98	63/M	4.5	III	1	Alive, 21	40-45,X,-Y,der(3)t(3;5)(p12~14;q14~21),-14[cp17]/88,idemx2[1]
99	64/M	1.5	IV	2	Alive (DP), 22	46-47,XY,der(3)t(3;5)(p13;q13~15),del(9)(p22)[cp29]/91-92,idemx2[cp4]/46,idem,i(8)(q10)[2]/46,XY[1]
100	54/F	6	I	2	NA	85-86,XXXX,+X,+X,-1,der(3)t(3;5)(p13;q22),-4,-4,-6,-8,-9,-14,-18[cp3]/79-83,idem,-X,-13,-22[cp10]/81-83,idem,-X,-13,-14,-22[cp5]

patients, 22 (21.1%) had metastases at the time of surgery, and in another 12 (11.5%), the disease progressed during the follow-up, leading to death from disease in 15 and 7 patients, respectively.

Tumor Size, T Stage, and Clinical Stage. The mean tumor size was 6.5 cm (median, 6 months; range, 1.5–16 cm). Tumor size was not significantly associated with overall survival. There were 52 pT_1,

Table 1 *Continued*

101	60/F	13	III	2	Alive, 20	48-49,XX,add(1)(p32),der(3)t(3;5)(p12;q31),+7,+12,-14,+16,+20,-21[cp4]/87-98,idemx2[cp8]/48-51,idem,dup(17)(q21q25)[cp2]/87-96,idemx2,dup(17)(q21q25)x2[cp6]/43-46,XX[cp12]
102	36/M	10	IV	3	DOTD, 2	45,XY,add(2)(q3?4),-3,der(4)t(3;4)(q21;q31)[3]/64-73<4n>,idemx2,-Y,-Y,del(1)(p31),-2,-2,add(5)(p12)x2,-7,-8,-8,-9,-9,add(11)(p15)x2,-12,-13,-13,-14,-14,-15,-15,-16,-17,-18,-18,-19,add(19)(q13),-21,-22,+mar1,+mar2[cp13]/46,XY[2]
103	68/F	6	III	1	NA	45,XX,der(1)t(1;8)(q12;q11),der(3)t(1;3)(q12;p21),-8[cp7]/47,XX,+7[1]
104	63/F	11	III	2	Alive, 12	46-51,X,-X,add(1)(q4?),+2,-3,add(4)(q3?),+5,+5,+5,+12,del(14)(q22q24),+16,-19,+mar1[cp23]
105	69/M	3.5	I	1	Alive, 3	40-45,X,-Y,-3,+5,add(9)(q22),der(11)t(3;11)(q13;q25),-14[cp46]/76-86,idemx2[cp3]
106	72/M	4	I	1	Alive, 11	45-53,X,-Y,+2,-3,add(3)(q13),+5,+7,del(11)(q14),+12,-16,+20,+3mar[cp11]/101,idemx2,+X,-12,-21,+2mar[1]/36-51,idem,i(8)(q10)[cp6]/45-51,idem,add(9)(p24),add(14)(q32)[cp6]
107	70/M	5.5	III	3	NA	68-74<4n>,XX,-Y,-Y,-1,-2,-3,-3,-4,-4,-5,der(6)t(3;6)(q11;q11)x2,-8,i(8)(q10),-9,-9,-10,-11,-13,-14,-15,-17,-18,+mar1[cp5]/71-73,idem,-14[cp3]/65-74,idem,-18[cp4]/67-72,idem,-16,-18[cp6]
108	63/F	4.5	I	2	Alive, 22	38-46,XX,del(3)(p14)[cp11]/46,XX[1]
109	71/M	4.5	I	1	Alive, 8	37-49,X,-Y,-3,+5,add(8)(q24),der(11)t(7;11)(q21;q14),+12,-14,+16,add(18)(q12),+20[cp28]/47-49,idem,+21[cp9]/47-49,idem,del(6)(q21)[cp2]/44-48,idem,+2[cp2]/48-49,idem,+2,add(21)(q22)[cp2]
110	70/F	9	IV	2	Alive (DP), 8	45-46,XX,der(3)t(3;5)(p13;q15),+7,-21[cp27]
111	62/M	8.8	II	2	NA	40-48,XY,del(1)(p34~35),der(3)t(3;5)(p14;q22),+der(3)t(3;5)(p14;q22),+12,-14[cp14]/93,idemx2,-8,-12,+mar[1]/46-47,idem,i(8)(q10)[cp2]
112	60/M	10	II	2	Alive, 3	38-44,X,-Y,+der(2)t(2;3)(p2;q2),-3,+del(7)(q22),-9,-14[cp4]/45,idem,+20[cp2]/46,XY[4]
113	69/M	12	IV	2	Alive, 3	45-46,XY,der(3)t(3;10)(p25;q11),del(4)(p11),+5,-7,add(8)(q24),-10,add(17)(q24),+mar[cp2]/61,idemx2,-Y,-Y,-1,-1,-del(4)(p11),-del(4)(p11),-9,-9,-10,-10,-12,-12,-13,-13,-13,-15,-15,-16,-16,-18,-18,-21,-21,-20,-20,-20,-20,-22,-22,-mar,-mar[1]/46-47,XY,+12[cp4]/42-47,XY,+7[cp3]/43-47,XY[cp30]
114	70/F	8	III	2	Alive, 3	40-44,XX,del(1)(p32),der(3)t(3;5)(p12-14;q14-22),-4,-8,-9,-14,add(21)(p11),+mar1[cp49]/86,idemx2[cp2]
115	61/F	9	IV	2	Alive, 1	35-45,XX,der(3)t(3;8)(p12;q11),-8[cp12]/35-43,idem,-14[cp4]/39-43,idem,-9,-14[cp3]/42-43,idem,-9,-14,+mar1[cp2]/35-44,idem,-1,add(9)(q34),-14,+mar1[cp5]/43-46,XX[cp9]
116	59/M	10	III	2	Alive, 14	44-46,der(3)t(3;5)(p12;q15),del(10)(q24)[cp2]/37-46,XY,idem,del(7)(q22),-del(10)(q24),+der(10)t(7;10)(q22;q24)[cp3]/46,idem,der(6)t(6;22)(q11;q11),del(7)(q22),-del(10)(q24),+der(10)t(7;10)(q22;q24)[2]/45-46,XY[cp3]/90,XX,-Y,-Y[1]
117	51/M	1.8	I	2	Alive, 9	69-80<4n>,XXYY,-1,-3,-3,-8,-8,-9,-10,-10,-13,-14,-16,-18,-18,-22[cp4]/60-67,idem,-1,-4, -6,-11,-12,-15,-17,-17,-21,-22[cp8]/64-65,idem,-1,-4,-4,-6,-11,-12,-15,-17,-17,-21,-22 [cp4]/57-64,idem,-1,-4,-4,-6,-9,-11,-12,-14,-15,-17,-17,-21,-22[cp11]/46-47,XY,+7[cp2]/43-47,XY[cp10]
118	70/F	11	II	2	Alive, 1	43-46,XX,der(3)t(3;5)(p12;q21)[cp17]/46,idem,del(5)(q15)[cp3]/43-46,XX[cp4]

13 pT_2, 50 pT_3 (6 pT_{3a}, 43 pT_{3b}, and 1 pT_{3c}), and 3 pT_4 tumors. Forty-nine (41.5%) patients had a tumor in clinical stage I, 12 (10.2%) in stage II, 34 (28.8%) in stage III, and 23 (19.5%) in stage IV. Among the 104 patients with available follow-up, patients with pT_1 or pT_2 tumors had a significantly prolonged survival compared with those with pT_3 or pT_4 tumors ($P = 6 \times 10^{-7}$). Likewise, patients in low clinical stage (stages I and II) had a prolonged survival (52 patients, 3 deaths; estimated probability for 5-year survival 0.95; 95% CI, 0.88–1.00), as opposed to patients in high clinical stage (stages III and IV; 52 patients, 19 deaths; estimated probability for 5-year survival of 0.44; 95% CI, 0.28–0.68). The differences in overall survival were highly significant ($P = 2 \times 10^{-6}$).

Histopathology and Grade of Malignancy. All 118 RCCs had purely clear cell cytomorphology, and 115 tumors had a predominant compact growth pattern. Three tumors consisted of clear cells with a mainly papillary or tubulo-papillary architecture (cases 51, 71, and 75), however, without foamy macrophages as otherwise commonly seen in papillary RCC. Forty-two (35.6%) tumors were grade of malignancy 1, 63 (53.4%) grade 2, and 12 (10.2%) grade 3. There was a strong association between clinical stage and grade of malignancy ($P = 0.0005$). Among the patients with available follow-up, patients with grade 1 tumors had the best outcome (36 patients, 2 deaths; estimated probability for 5-year survival, 0.94; 95% CI, 0.86–1.00), whereas patients with grade 2 tumors (59 patients, 13 deaths; estimated probability for 5-year survival of 0.60; 95% CI, 0.44–0.82), and patients with grade 3 tumors (9 patients, 7 deaths; estimated probability for 5-year survival, 0.26; 95% CI, 0.08–0.85) had a distinctly worse overall survival. According to the three-sample log-rank test, the differences in overall survival were highly significant ($P = 2 \times 10^{-9}$).

Cytogenetics. All 118 clear cell RCCs had aberrant karyotypes with an average of seven aberrations/tumor (median, 5; range, 1–27; Table 1). Low numbers of karyotypic changes (<4) were associated with low grade of malignancy ($P = 0.0025$); however, the difference in overall survival was not significant. Intratumor karyotypic heterogeneity detected as the presence of more than one clone within a tumor was observed in 67 cases; the different clones within the same tumor (range, 2–8) were related except in case 3. Considering the most complex clone in a tumor, the chromosome number was hypodiploid in 48, pseudodiploid in 27, hyperdiploid in 21, and polyploid in 22 tumors. Polyploidy was correlated with a higher grade of malignancy ($P = 0.034$); however, none of the different ploidy levels were significantly associated with overall survival.

The most common stem-line change was loss of chromosome 3p,

which occurred in 116 (98.3%) cases. Thirty-six (31%) were unbalanced translocations der(3)t(3;5)(p11–p22;q13–q31), resulting in loss of chromosome 3p (smallest overlapping deletion, 3p22–pter) and duplication of 5q (smallest overlapping duplication, 5q31–qter; Fig. 1*A*). This distinct chromosomal translocation represented the single most frequent structural aberration in clear cell RCC. Another 48 (41.4%) were structural aberrations affecting 3p other than translocation 3;5 and included 19 terminal deletions, 11 add(3), 5 der(3)t(3;8), 2 der(3)t(2;3), 2 der(3)t(3;7), 5 nonrecurrent translocations involving chromosomes 1, 6, 10, 13, and 14, respectively; one ins(3), leading to interstitial loss of 3p22–p24 sequences, 1 dic(3;15), and 2 ring chromosomes. In all but one case (case 113) of the 84 clear cell RCCs with structural aberrations of 3p, the breakpoints were determined between bands 3p11–p22 (smallest overlapping deletion, 3p22–pter; Fig. 1*B*). The remaining 32 (27.6%) aberrations were monosomies or numerical losses of chromosome 3, of which 16 were combined with unbalanced translocations involving breakpoints at 3q11–q21, including 5 der(4)t(3;4), four der(6)t(3;6), 2 der(1)t(1;3), and 5 nonrecurrent translocations involving chromosomes 2, 7, 11, 20, and 22, respectively. There was a significant association between der(3)t(3;5) and overall survival, in that patients with this translocation (32 patients, 1 death; estimated probability for 5-year survival, 0.94; 95% CI, 0.84–1.0) had a significantly better outcome than those without der(3)t(3;5) (72 patients, 21 deaths; estimated probability for 5-year survival, 0.64; 95% CI, 0.52–0.79; $P = 0.008$). Also among the 34 patients with distant metastasis at the time of surgery or disease progression during follow-up, death of disease occurred in 1 of 8 (12.5%) patients with der(3)t(3;5) compared with 21 of 26 (80.8%) patients without this aberration. Fisher's exact test showed no significant association of der(3)t(3;5) with tumor size, low *versus* high TNM stage, low *versus* high clinical stage, and grade of malignancy.

Gain of 5q31–qter was the second most frequent stem-line aberration, occurring in 67 (56.8%) RCCs (61 stem-line aberrations, 6 side-line aberrations), resulting from either polysomies (26 cases; 20 stem-line aberrations, 6 side-line aberrations), structural rearrangements involving chromosome 5 with breakpoints ranging between bands 5q11–5q31 (44 cases: 36 der(3)t(3;5), 3 der(6)t(5;6), 2 der(1)t(1;5), 1 der(2)t(2;5), and 1 der(4)t(4;5) all as stem-line aberrations, 1 der(14)t(5;14) as side-line aberration; Fig. 1*B*), or both (3 cases). Patients with gain of 5q31–qter in their stem-lines, regardless of cytogenetic origin, had a prolonged overall survival compared with patients without gain of 5q ($P = 0.001$; Fig. 2). Considering only patients in high-clinical stage, those without gain of 5q had a distinctly worse overall survival (23 patients, 15 deaths; estimated probability for 5-year survival, 0.16; 95% CI, 0.05–0.54), whereas patients with a gain of 5q had far better survival rates (29 patients, 4 deaths; estimated probability for 5-year survival, 0.70; 95% CI, 0.47–1.00). The difference in overall survival was highly significant ($P = 3 \times 10^{-5}$; Fig. 3). Again, there was no strong association of gain of 5q with any of the classical clinicopathological variables.

Losses of chromosome 14q were present in 74 (61.4%) RCCs,

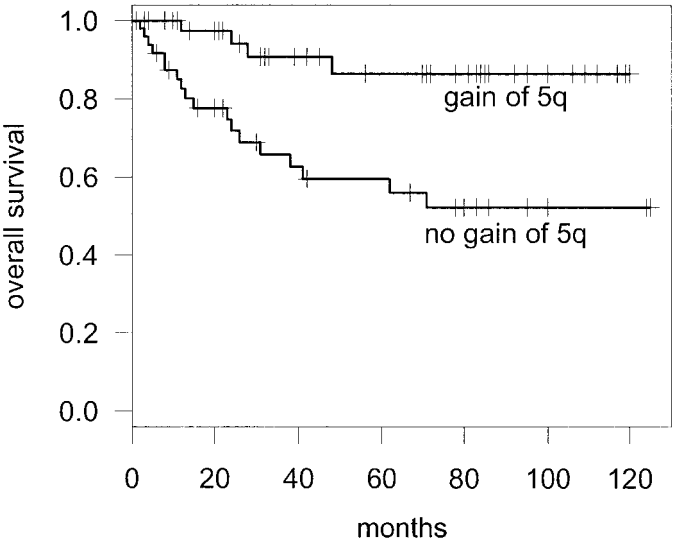

Fig. 2. Kaplan-Meier plot of overall survival of RCC patients with 5q gain (52 patients, including 32 patients with der(3)t(3;5), 4 deaths) and without 5q gain (52 patients, 18 deaths; $P = 0.001$, log-rank test).

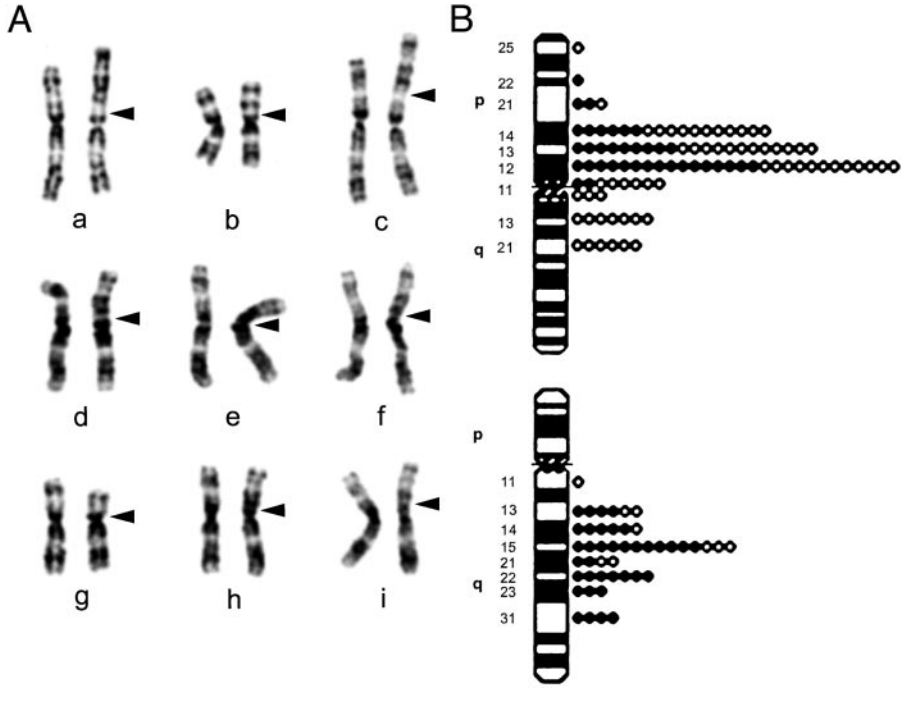

Fig. 1. Partial karyotypes of nine different tumors showing unbalanced chromosomal translocation der(3)t(3;5) in cases 6 (*a*), 12 (*b*), 20 (*c*), 59 (*d*), 62 (*e*), 81 (*f*), 58 (*g*), 87 (*h*), and 100 (*i*) with variations of chromosome breaks at 3p and 5q (*A*). Chromosome breakpoints affecting chromosome 3 (*above*) and chromosome 5 (*below*) in 118 cases of primary clear cell RCCs are shown. *B:* ●, chromosome break in der(;3)t(3;5); ○, chromosome break in structural aberrations other than der(3)t(3;5).

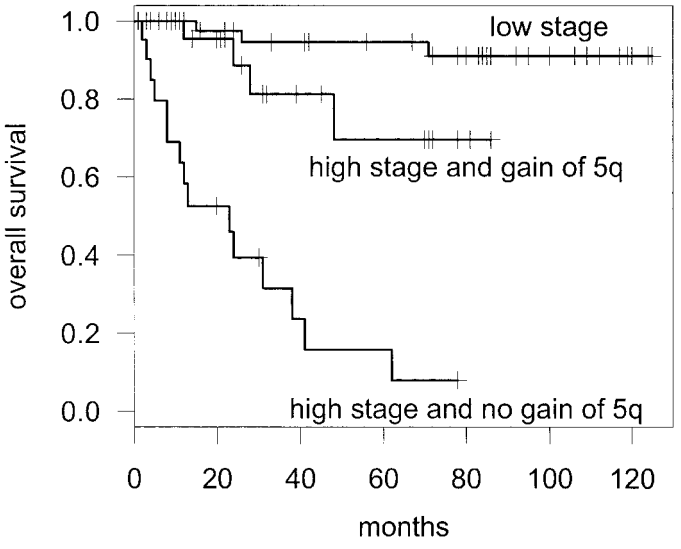

Fig. 3. Kaplan-Meier plot of overall survival of low-clinical stage RCC patients (52 patients, 3 deaths) *versus* high clinical stage RCC patients with 5q gain (25 patients, including 19 with der(3)t(3;5), 4 deaths) and without 5q gain (23 patients; 15 deaths).

resulting from either monosomies (65 cases; 44 stem-line aberrations, 21 side-line aberrations) or structural rearrangements involving 14q (9 cases; 2 stem-line aberrations, 7 side-line aberrations). Further recurrent aberrations occurring in at least 10% of the 118 karyotyped RCCs included −8p/−8 (33.1%), −9/−9p (23.7%), −6q/−6 (23.7%), −Y (41.7% of male patients), −1p/−1 (18.6%), +7/+7q (18.6%), +2/+2q (14.4%), −4q/−4 (14.4%), −18/−18q (14.4%), +12 (13.6%), +8q/+8 (12.7%), −10/−10q (11%), and +20 (10.2%). As a tendency, partial chromosomal net changes including −8p, −6q, −4q, +5q, +2q, and −9q were more prevalent in hypodiploid or pseudodiploid tumors, whereas polysomies, in particular +5, +12, +20, +16, +7, and +2, dominated in the hyperdiploid tumors. Tumors with polyploid stem-line chromosome numbers frequently had numerical losses, such as −6, −8, −18, −9, −10, −17, and −4, respectively. Correlation with clinicopathological variables and overall survival was calculated for all common autosomal aberrations. Loss of chromosome 1/1p (22 patients) was associated with larger tumors ($P = 0.04$). Losses of 4q/4 (13 patients) and/or 15/15q (8 patients) were associated with higher N stage ($P = 0.04$ and $P = 0.006$, respectively). Another correlation was found between loss of chromosome 9/9p (25 patients) and metastasis at the time of surgery ($P = 0.006$), whereas all 13 patients with gain of chromosome 2/2q were free of distant metastasis at the time of surgery ($P = 0.04$). Loss of chromosome 18/18p (14 patients) was associated with higher grade of malignancy ($P = 0.02$).

Discussion

The aim of the present study was to evaluate the prognostic significance of cytogenetic aberrations for a large series of patients with primary clear cell RCCs. As suspected, within our patient population, clinical stage and grade of malignancy were the strongest predictors of likely outcome, in accordance with most previous series (6, 7). Furthermore, this study shows that specific cytogenetic aberrations can add to the prognostic value of classical clinicopathological indicators of prognosis. Although loss of chromosome segment 3p14–pter and der(3)t(3;5) leading to simultaneous duplication of 5q have long been known to be characteristic aberrations in clear cell RCCs (4, 5), this study is the first to indicate that gain of 5q31–qter might identify a clinically favorable subgroup of clear cell RCCs with prolonged overall survival. It is tempting to assume that the favorable prognosis in patients with gain of 5q31–qter is related to a less advanced disease stage, but there was no association of gain of 5q or der(3)t(3;5) with any of the classical clinicopathological variables, including low *versus* high clinical stage, and grade of malignancy. The difference in survival appeared to be uninfluenced by advanced disease; among the patients in high-clinical stage (stages III and IV), those with a gain of 5q had survival rates comparable with those in low clinical stage (stages I and II), whereas patients without gain of 5q had a distinctly worse overall survival. Given the considerable variation of breakpoints at 5q, functional gain of the minimal duplicated region 5q31–qter appears to be the most critical genetic determinant of favorable clinical outcome. A recent RFLP study has shown a breakpoint cluster between the *APC* and *MCC* genes at distal 5q21 and identified two distinct interstitial duplications of DNA sequences distal to the *APC/MCC* genes, one at the 5q22 region (including the loci *D5S659*, *D5S1720*, and *w2005*) and another at the 5q31 region (including the loci *D5S816*, *D5S476*, and *D5S1480*), harboring the nonfunctional α-catenin pseudogene *CTNNAP1* and the functional α-catenin gene *CTNNA1*, respectively (14). Alterations in expression of cell-cell adhesion molecules have been implicated in tumor progression in a number of carcinomas, and decreased α-catenin expression detected by immunohistochemistry has been associated with poor prognosis in patients with localized RCC (15). Further molecular studies have to be awaited to confirm a possible functional gain of *CTNNA1* as a likely determinant of favorable prognosis in clear cell RCC.

Among the additional recurrent cytogenetic abnormalities, none were significantly associated with overall survival; however, there were a few associations with clinicopathological variables: loss of chromosome 1/1p with larger tumor size; losses of 4q/4 and/or 15/15q with higher N stage; gain of chromosome 2/2q with absence of distant metastasis at the time of surgery; loss of chromosome 18/18p; higher degree of cytogenetic complexity; and polyploid chromosome numbers with higher grade of malignancy. Given the multiple testing and the comparably low number of informative cases, these associations have to be interpreted with caution; however, they may indicate notable trends, and some associations support previous findings. The observed correlation between tumors with four and more aberrations and higher grade of malignancy is consistent with the assumption that a net accumulation of genetic events is responsible for tumor progression in RCC. Likewise, other reports have implicated a higher degree of cytogenetic complexity with worse prognosis (10, 11); however, in our study, there was no significant relationship between the total number of aberrations and overall survival. The association between aneuploid polyploidy and grade of malignancy is analogous to observations obtained by others (16). The strong correlation between loss of chromosome 9/9p and metastasis at the time of surgery seen in this study confirms a role of a tumor suppressor gene on 9p involved in RCC progression. This finding is in agreement with other reports suggesting an association of 9p deletions and progression of both clear cell and papillary RCCs (10, 17). Although several candidate tumor suppressor genes at 9p21, including *CDKN2A*, *CDKN2B*, and *MTAP*, have been implicated as possible targets of inactivation in a variety of human neoplasms (18, 19), recent data suggest the existence of one or several other tumor suppressor genes outside the 9p21 region as potential targets of the observed deletions. This might also apply for RCCs, because loss of heterozygosity was most frequently observed proximal to *CDKN2A* at 9p13 (17), and potential inactivation of *CDKN2A* by homozygous deletions, rearrangements, or point mutations was found to be rare in RCC (20).

The data highlight that gain of 5q31–qter might be an independent prognostic indicator that identifies a clinically favorable cytogenetic variant of clear cell RCC, providing the basis for a clinically useful subdivision within this histologically defined tumor category. Even-

tually, classification into genetically and clinically distinct subgroups may be helpful in predicting likely outcome and designing appropriate therapeutic strategies. Furthermore, clinical trials testing cancer therapeutics, such as immunization or immunomodulatory treatment of metastatic RCC disease, will benefit from the definition of homogeneous patient populations improving the likelihood of observing efficacy of a specific therapeutic regimen. In addition, several cytogenetic abnormalities might have prognostic significance in patients with clear cell RCC; in particular, loss of chromosome 9/9p might prove a relevant marker for RCC progression and metastasis.

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