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Integration of Recurrent Somatic Mutations with Clinical Outcomes: A Pooled Analysis of 1049 Patients with Clear Cell Renal Cell Carcinoma

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Abstract

Background—Analyses of associations between clinicopathologic outcomes and recurrent somatic mutations in clear cell renal cell carcinoma (ccRCC) have been limited to individual cohorts.

Objective—To define clinicopathologic associations between specific mutations and ccRCC disease characteristics.

Design, setting, and participants—DNA sequencing data were pooled from three collaborative genomic cohorts (n = 754) and our institutional database (n = 295). All patients had clinical data and identification of somatic mutations from their primary tumors.

Outcome measurements and statistical analysis—Analysis of gene mutations for associations with maximal tumor size (linear regression) and pathologic stage (logistic regression). Cancer-specific survival (CSS) and recurrence-free survival (RFS) were calculated using competing risks methods. Analyses were adjusted for cohort site, and results were adjusted for multiple testing (q value). Relevant genes were used in multivariable models that included confounding variables and the validated Mayo Clinic Stage, Size, Grade, and Necrosis (SSIGN) score.

Results and limitations—Association with tumor size was found for mutations in *BAP1* (q = 0.013). No mutations were found to be associated with stage after adjusted analysis. Mutations in *BAP1* (q = 0.004) and *TP53* (q = 0.001) were associated with decreased CSS in a multivariable model; only *TP53* (q = 0.005) remained significant when SSIGN score was included. *SETD2* mutations (q = 0.047) were associated with decreased RFS in multivariable models, including models with SSIGN score.

Conclusions—In >1000 patients with ccRCC, pooled analysis and multivariable modeling demonstrated that three mutated genes have statistically significant associations with poor clinical outcomes. This included the more commonly mutated *BAP1* and *SETD2* and the less frequently mutated *TP53*. After adjustment for clinical confounders, mutations of *TP53* and *SETD2* were associated with decreased CSS and RFS, respectively.

Patient summary—Using rigorous statistical methods, this study affirmed that certain mutations in clear cell renal cell carcinomamay portend inferior survival and an increased risk of recurrence.

Keywords

Mortality; Mutation; Prognosis; Renal cell carcinoma; Sequencing analysis; DNA

1. Introduction

Despite recent advances in surgical and systemic treatments, renal cell carcinoma (RCC) remains the most lethal urologic malignancy, accounting for about 3% of all human cancers.

Clear cell RCC (ccRCC) is the most common and aggressive histologic subtype [1]. The advent of next-generation sequencing technology has resulted in a fundamental shift in the understanding and potential treatment of ccRCC. Large-scale efforts by cooperative groups like The Cancer Genome Atlas [2], the International Cancer Genome Consortium [3], and the University of Tokyo [4] helped define the genomic landscape of ccRCC [5] and identified several recurrently mutated genes [6–8].

Observations and interpretations of the genetic landscape of ccRCC have been limited by several constraints. Most of the reports in this arena are based on relatively small patient cohorts with similar pathologic stages [2–4,9,10]. This, coupled with the low frequency of some mutations, has resulted in multivariable analyses being underpowered.

In this study, we performed comprehensive analyses of pooled publicly available cohorts, along with our institutional cohort, to identify associations between relevant mutations in ccRCC and clinicopathologic outcomes while controlling for known prognostic variables.

2. Methods

2.1. Patient selection

All patients included in the study signed informed consent at their respective institutions allowing for genomic testing. On approval by the institutional review board at Memorial Sloan Kettering Cancer Center (MSKCC), we searched our institutional kidney cancer database and identified 348 patients with ccRCC with prospectively collected genomic and clinical data between 2001 and 2015 (MSKCC cohort). Patients were excluded from this study if sequencing had not been performed on their primary tumors ($n = 53$), leaving 295 patients available for analysis. This included patients ($n = 185$ [62.7%]) who had targeted sequencing of five genes and had been previously described [11] but now had >3 yr longer follow-up. Of these 185 patients, 54 (29.2%) had next-generation sequencing performed, which was used in place of their targeted sequencing results for this analysis. One hundred ten patients (37.3%) in the MSKCC cohort have not been previously described. A majority of patient samples ($n = 157$ [53.2%]) were analyzed using MSK-IMPACT (MSKCC Integrated Mutation Profiling of Actionable Cancer Targets), a hybridization-based exon capture assay of select introns and commonly altered oncogenes and tumor suppressor genes [12]. The panel of targeted genes is listed in Supplement 1. Details of our genomic pipeline have been previously published [12]. For 35 patients, sequencing data were obtained using MSK-IMPACT performed on dissociated cells from their primary kidney tumors after an average of 1.7 passages. Remaining samples were analyzed using Sanger ($n = 131$ [44.4%]) and whole-genome ($n = 7$ [2.4%]) sequencing.

The second cohort in this study (public cohort) consisted of three previously published cohorts of patients with ccRCC [2–4], with documentation of appropriate ethics and consent for all study participants. Details of these study populations may be found in Supplement 2.

2.2. Statistical analysis

Data from a total of 1049 patients were available for analysis. A panel of 14 genes was chosen for analysis based on previously published works focusing on the clinical relations of

significantly mutated genes across all cohorts. All patients had data available on four mutated genes (*VHL*, *PBRM1*, *SETD2*, and *BAP1*). Depending on the original study parameters, the 10 remaining genes had varying levels of data missing. To account for differences in patient characteristics and mutation frequencies across cohorts, all analyses were adjusted for cohort by including cohort as a categorical variable in all regression models. Linear regression analysis was used to assess the association between maximum pathologic tumor diameter and genetic mutations. Logistic regression estimated the association between gene mutations and American Joint Committee on Cancer (AJCC) stage [13]. We examined two ways of categorizing AJCC stage: (1) dichotomized as stages I/II/III versus stage IV and (2) categorized as stages I/II versus stage III versus stage IV.

Follow-up time was calculated from date of nephrectomy. Recurrence was determined according to previously described methods among the patients from the public cohort [2–4]. Date of recurrence was defined as date of pathologic confirmation of diagnosis, that is, biopsy. For patients with no pathology-confirmed recurrence, we used the date of the radiologic examination at which recurrence was diagnosed. Patients with documented recurrence who were reported as deceased on last follow-up and those with stage IV disease who were deceased without a listed cause were considered to have died from their disease. We used competing risks methods to estimate recurrence-free survival (RFS) and cancer-specific survival (CSS), treating death without recurrence and death from other causes as competing events, respectively.

To control for potential confounding in multivariable models, two sets of covariates were determined a priori. The first set, the base set, included age at diagnosis, sex, AJCC stage, and Fuhrman nuclear grade. The second set, the Mayo Clinic Stage, Size, Grade, and Necrosis (SSIGN) set, included age at diagnosis, sex, and SSIGN score as a continuous variable. The SSIGN score [14] is a validated prognostic model that is widely used to control for confounding in analytic models for RCC prognosis; because a SSIGN score was not available for all patients under study, it was treated as a secondary analysis.

All q values reported in this paper are the p values adjusted for multiple testing using the Benjamini-Hochberg false discovery rate method. A q value <0.05 was considered statistically significant. All statistical analyses were conducted using R software v.3.1.1 (R Core Development Team, Vienna, Austria), including the survival and *cmprsk* packages.

3. Results

3.1. Demographic characteristics

Patient and disease characteristics, including gene mutation frequencies, are described in Table 1. The composite cohort (including both MSKCC and public cohorts) displayed classic recurrence and survival patterns for ccRCC.

3.2. Tumor size

Data on tumor size were available for 1045 patients. Tests for the association between each gene and tumor size were conducted using the available sample size for each respective gene. In analyses adjusted only for cohort, mutations in *BAP1* (median: 8 vs 5.2 cm; p =

0.001) and *PTEN* (median: 8 vs 5.9 cm; $p = 0.026$) were significantly associated with larger median tumor diameter. After adjustment for multiple comparisons, mutations in *BAP1* remained significantly associated with larger tumor size ($q = 0.013$) (Supplementary Table 1).

We further tested the difference in mutation frequency among tumors smaller than 4 cm, stratifying between those with pathologic (p) T1 classification and those with pT3 classification for the five most recurrently mutated genes in the composite cohort (*VHL*, *PBRM1*, *SETD2*, *BAP1*, and *KDM5C*). On analyses adjusted only for cohort, we found *PBRM1* ($p = 0.021$) and *KDM5C* ($p = 0.040$) to be associated with pT3 classification; after controlling for multiple testing, neither gene retained significance ($q = 0.099$ for both) (Supplementary Table 4).

3.3. Disease stage

Analyses for the association between each gene and stage were conducted using the available sample size for each respective gene. On analyses adjusted only for cohort, several genes were found to be associated with higher-stage disease (Supplementary Table 1 and 2); after adjustment for multiple comparisons, no genes were significantly associated with a specific stage in either analysis. Of note, mutations in *SETD2*, *PTEN*, and *BAP1* did show a trend toward significance for association with higher AJCC stages when comparing stage I/II versus stage III versus stage IV, with q values of 0.071, 0.084, and 0.088, respectively.

3.4. Cancer-specific survival

Investigation of the association between each mutated gene and CSS was conducted using the available sample size for each respective gene. On analyses adjusted only for cohort, we found that mutations in *BAP1* ($n = 1049$ for analysis; hazard ratio [HR]: 2.10; 95% confidence interval [CI]: 1.44–3.04; $q < 0.001$) and *TP53* ($n = 784$ for analysis; HR: 2.63; 95% CI: 1.36–5.08; $q = 0.028$) were significantly associated with increased risk of death from cancer after adjustment for multiple comparisons using competing risks regression (Fig. 1).

We consequently incorporated *BAP1* and *TP53* into multivariable competing risks regression models using the two sets of adjustment covariates described in the statistical methods. In the first model, which was adjusted for the base set of variables as well as cohort ($n = 733$ for analysis), we found both *BAP1* ($q = 0.004$) and *TP53* ($q = 0.001$) mutations to be significantly associated with decreased CSS (Supplementary Table 5). In the second model, which was adjusted for the SSIGN set of variables as well as cohort ($n = 554$ for analysis), we found that only *TP53* mutations were significantly associated with decreased CSS (Table 2). To assess the presence of sarcomatoid features as a possible confounding variable, we included this in a model with the base set of variables. We still found *TP53* and *BAP1* mutations to be associated with inferior CSS (Table 3).

3.5. Recurrence-free survival

After excluding the data of patients with AJCC stage IV disease, we looked for an association between the selected genes and RFS in 860 patients (depending on the available

sample size for each respective gene). On analyses adjusted only for cohort, we found that mutations in *SETD2* (HR: 1.89; 95% CI: 1.26–2.83; $q = 0.007$), *KDM5C* (HR: 2.02; 95% CI: 1.29–3.18; $q = 0.007$), *TP53* (HR: 3.12; 95% CI: 1.39–7.00; $q = 0.017$), *PTEN* (HR: 3.78; 95% CI: 1.94–7.37; $q < 0.001$), and *TSC1* (HR: 4.15; 95% CI: 2.04–8.41; $q < 0.001$) were significantly associated with increased risk of recurrence (Supplementary Table 6).

Our multivariable models included *SETD2* and *KDM5C* but not *PTEN*, *TSC1*, or initially *TP53* because of their lower overall mutation frequency (4.08%, 1.66%, and 3.44%, respectively) and the risk of overfitting. In the multivariable model adjusted for the base set of variables as well as cohort ($n = 733$ for analysis), only *SETD2* showed statistically significant association with increased risk of recurrence (HR: 1.60; 95% CI: 1.04–2.47; $q = 0.033$) (Supplementary Table 7). In the multivariable model adjusted for the SSIGN set of variables as well as cohort ($n = 539$ for analysis), *SETD2* continued to demonstrate significance (Table 4). To assess the specific correlation of *TP53* with RFS, it was included with *SETD2* in two additional multivariable models with both sets of variables. We saw no significance for either gene in these models (Supplementary Table 8 and 9).

4. Discussion

This pooled analysis of data from >1000 patients with ccRCC, with genomic characterization of their primary tumors, allowed us to elicit the clinical implications of several recurrently mutated genes using more rigorous multivariable statistical modeling. We identified several somatic mutations that act as an inflection point in the pathogenesis of ccRCC. On univariable analysis, adjusted for cohort, we found six genes that were associated with clinical outcomes: *BAP1*, *SETD2*, *KDM5C*, *PTEN*, *TP53*, and *TSC1*. Three of these genes, *BAP1*, *TP53*, and *SETD2*, continued to show significance in multivariable modeling.

Previously published studies have reported a more aggressive clinical course and worse prognosis in patients who have ccRCC with *BAP1* mutations [11,15]. In this investigation, *BAP1* mutations were associated with increased risk of cancer-specific death, but the association did not retain significance on inclusion of the SSIGN score in a multivariable model. One reason for these outcomes may be the association of *BAP1* mutations with larger tumors and with histologic grade 4 disease compared with grades 1–3 ($p < 0.001$; data not shown). Both of these pathologic variables are captured in the SSIGN score calculation, and *BAP1* mutations may be surrogates for these variables.

Notably, we found no association between *BAP1* mutations and decreased RFS, even on univariable analysis with adjustment for cohort site. A *BAP1* mutation may encourage tumor cell growth, but unlike other mutations (eg, *SETD2* and *TP53* mutations), it may not facilitate dissemination of tumor cells [8,16].

While generally rare, mutations in *TP53* were predictive of several adverse clinical outcomes, including decreased CSS and RFS, in this study. Strikingly, *TP53* mutations maintained their association with increased risk of cancer-specific death even when controlling for the validated SSIGN score. The association with lower RFS was not found to

be significant in our multivariable models, which may highlight the importance of this mutation in those with stage IV disease. Other investigators have reported the enrichment of *TP53* mutations in patients with metastatic disease [17] and in the aggressive sarcomatoid variant of ccRCC [18]. For the latter, we found *TP53* to remain significant even when including this pathologic feature in a multivariable model.

Previous studies have associated *SETD2* with advanced stage, tumor invasiveness, and development of metastatic disease [4,19,20]. In this study, *SETD2* was associated with recurrence in models that were adjusted for both sets of variables.

A number of previous investigations have sought to identify prognostic biomarkers in ccRCC, and several studies focused on recurrently mutated genes along with cytogenetic alterations [10,11,15,21–24] in isolated cohorts. Many of the molecular biomarkers assessed in previous studies offered little or no advantage over more traditional pathologic and clinical variables in the prediction of clinical outcomes [11,21]. This may be because some somatic aberrations act as a proxy for traditional pathologic and clinical variables.

The clinical application of molecular markers spans the full spectrum of ccRCC treatment and management. There is potential for molecular markers obtained from biopsied tissue to aid in the selection of patients for active surveillance and also from surgical specimens for the development of risk-adjusted postoperative follow-up. Furthermore, assessment of the mutational status of the genes reported in this study in the metastatic setting may provide guidance on precision systemic therapies, as was recently reported in an analysis of the RECORD-3 cohort [25]. We believe these areas to be prime for directed clinical studies.

4.1. Limitations

A noted limitation of this study is that all sequenced samples were derived from a single site of each patient's primary tumor. Tumor-variant allele frequency of each mutation in each patient was not evaluated in this study. Previous works from our group and others have highlighted the intratumoral heterogeneity in ccRCC. This is an inherent limitation in most previously published series in this arena, in both primary and metastatic tumor tissue [2–6,8,9,17,26]. Some balance between multiple sampling and clinical benefit must be considered. Pooling of multiple cohorts that use different sequencing platforms has an inherent risk of possibly overestimating or underestimating the true frequency of mutations, especially when compared with each other. The different patient cohorts demonstrated heterogeneity in both patient characteristics (eg, pathological stage) and frequency of gene mutations (eg, *VHL* mutations), and we controlled for this by adjusting for cohort in all statistical analyses.

5. Conclusions

In this study of >1000 patients with ccRCC, pooled analysis and multivariable modeling demonstrated that three recurrently mutated genes, *BAP1*, *SETD2*, and *TP53*, have statistically significant associations with poor clinical outcomes. After adjustment for important clinical confounders, mutations of *TP53* and *SETD2* were associated with decreased CSS and RFS, respectively.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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References

1. Global Burden of Disease Cancer Collaboration. The global burden of cancer 2013. JAMA Oncol. 2015; 1:505–27. [PubMed: 26181261]
2. Cancer Genome Atlas Research Network. Comprehensive molecular characterization of clear cell renal cell carcinoma. Nature. 2013; 499:43–9. [PubMed: 23792563]
3. Scelo G, Riazalhosseini Y, Greger L, et al. Variation in genomic landscape of clear cell renal cell carcinoma across Europe. Nat Commun. 2014; 5:5135. [PubMed: 25351205]
4. Sato Y, Yoshizato T, Shiraishi Y, et al. Integrated molecular analysis of clear-cell renal cell carcinoma. Nat Genet. 2013; 45:860–7. [PubMed: 23797736]
5. Guo G, Gui Y, Gao S, et al. Frequent mutations of genes encoding ubiquitin-mediated proteolysis pathway components in clear cell renal cell carcinoma. Nat Genet. 2012; 44:17–9.
6. Voss MH, Hakimi AA, Pham CG, et al. Tumor genetic analyses of patients with metastatic renal cell carcinoma and extended benefit from mTOR inhibitor therapy. Clin Cancer Res. 2014; 20:1955–64. [PubMed: 24622468]
7. Dalgliesh GL, Furge K, Greenman C, et al. Systematic sequencing of renal carcinoma reveals inactivation of histone modifying genes. Nature. 2010; 463:360–3. [PubMed: 20054297]
8. Peña-Llopis S, Vega-Rubín-de-Celis S, Liao A, et al. BAP1 loss defines a new class of renal cell carcinoma. Nat Genet. 2012; 44:751–9. [PubMed: 22683710]
9. Kapur P, Peña-Llopis S, Christie A, et al. Effects on survival of *BAP1* and *PBRM1* mutations in sporadic clear-cell renal-cell carcinoma: a retrospective analysis with independent validation. Lancet Oncol. 2013; 14:159–67. [PubMed: 23333114]
10. Pastore A, Jurinovic V, Kridel R, et al. Integration of gene mutations in risk prognostication for patients receiving first-line immunochemotherapy for follicular lymphoma: a retrospective analysis of a prospective clinical trial and validation in a population-based registry. Lancet Oncol. 2015; 16:1111–22. [PubMed: 26256760]
11. Hakimi AA, Chen YB, Wren J, et al. Clinical and pathologic impact of select chromatin-modulating tumor suppressors in clear cell renal cell carcinoma. Eur Urol. 2013; 63:848–54. [PubMed: 23036577]
12. Cheng DT, Mitchell TN, Zehir A, et al. Memorial Sloan Kettering-Integrated Mutation Profiling of Actionable Cancer Targets (MSK-IMPACT): a hybridization capture-based next-generation sequencing clinical assay for solid tumor molecular oncology. J Mol Diagn. 2015; 17:251–64. [PubMed: 25801821]
13. Edge, S.Byrd, DR.Compton, CC.Fritz, AG.Greene, FL., Trotti, A., editors. AJCC cancer staging manual. 7. New York, NY: Springer; 2010.
14. Frank I, Blute ML, Cheville JC, Lohse CM, Weaver AL, Zincke H. An outcome prediction model for patients with clear cell renal cell carcinoma treated with radical nephrectomy based on tumor stage, size, grade and necrosis: the SSIGN score. J Urol. 2002; 168:2395–400. [PubMed: 12441925]
15. Joseph RW, Kapur P, Serie DJ, et al. Clear cell renal cell carcinoma subtypes identified by BAP1 and PBRM1 expression. J Urol. 2015; 195:180–7. [PubMed: 26300218]
16. Ventii KH, Devi NS, Friedrich KL, et al. BRCA1-associated protein-1 is a tumor suppressor that requires deubiquitinating activity and nuclear localization. Cancer Res. 2008; 68:6953–62. [PubMed: 18757409]

17. Gerlinger M, Horswell S, Larkin J, et al. Genomic architecture and evolution of clear cell renal cell carcinomas defined by multiregion sequencing. Nat Genet. 2014; 46:225–33. [PubMed: 24487277]
18. Bi M, Zhao S, Said JW, et al. Genomic characterization of sarcomatoid transformation in clear cell renal cell carcinoma. Proc Natl Acad Sci U S A. 2016; 113:2170–5. [PubMed: 26864202]
19. Hakimi AA, Ostrovnaya I, Reva B, et al. ccRCC Cancer Genome Atlas (KIRC TCGA) Research Network Investigators. Adverse outcomes in clear cell renal cell carcinoma with mutations of 3p21 epigenetic regulators *BAP1* and *SETD2*: a report by MSKCC and the KIRC TCGA Research Network. Clin Cancer Res. 2013; 19:3259–67. [PubMed: 23620406]
20. Ho TH, Park IY, Zhao H, et al. High-resolution profiling of histone h3 lysine 36 trimethylation in metastatic renal cell carcinoma. Oncogene. 2016; 35:1565–74. [PubMed: 26073078]
21. Hakimi AA, Mano R, Ciriello G, et al. Impact of recurrent copy number alterations and cancer gene mutations on the predictive accuracy of prognostic models in clear cell renal cell carcinoma. J Urol. 2014; 192:24–9. [PubMed: 24518768]
22. Klatte T, Kroeger N, Rampersaud EN, et al. Gain of chromosome 8q is associated with metastases and poor survival of patients with clear cell renal cell carcinoma. Cancer. 2012; 118:5777–82. [PubMed: 22605478]
23. Klatte T, Rao PN, de Martino M, et al. Cytogenetic profile predicts prognosis of patients with clear cell renal cell carcinoma. J Clin Oncol. 2009; 27:746–53. [PubMed: 19124809]
24. Ricketts CJ, Linehan WM. Gender specific mutation incidence and survival associations in clear cell renal cell carcinoma (CCRCC). PLoS One. 2015; 10:e0140257. [PubMed: 26484545]
25. Hsieh, JJ., Wang, P., Chen, Y., et al. Differential overall survival (OS) results in RECORD-3 study based on three distinct mRCC molecular subgroups classified by BAP1 and/or PBRM1 mutations. Presented at: Genitourinary Cancers Symposium; January 9, 2016; San Francisco, CA.
26. da Costa WH, Rezende M, Carneiro FC, et al. Polybromo-1 (PBRM1), a SWI/SNF complex subunit is a prognostic marker in clear cell renal cell carcinoma. BJU Int. 2014; 113:E157–63. [PubMed: 24053427]

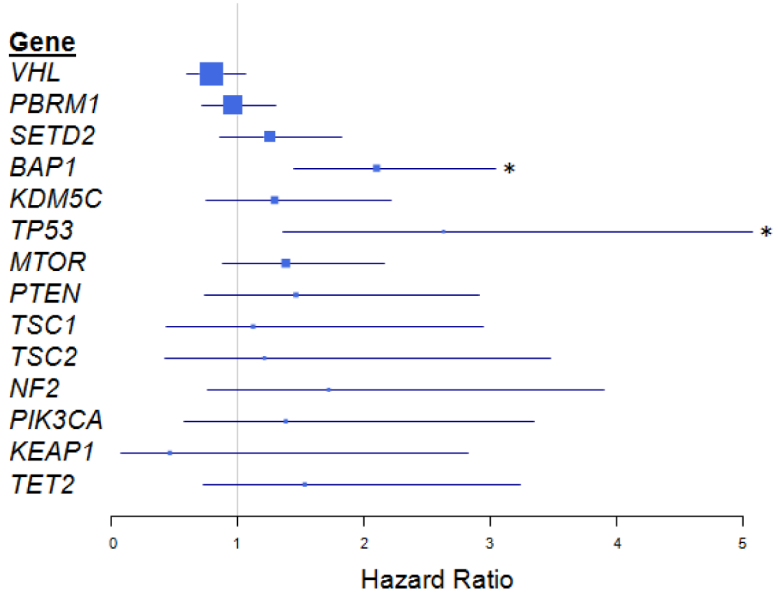

Fig. 1.

Forest plot of associations between gene mutations and cancer-specific survival, adjusted for study site.

Table 1

Patient characteristics for composite patient cohort

Characteristic	Composite cohort, *n* = 1049
Age at diagnosis, yr, median (range)	61 (21–91)
Maximum tumor dimension, cm, median (range)	5.5 (1–25)
BMI, kg/m^2, median (range)	28.9 (12.7–69.7)
SSIGN score, median (range)	4 (0–15)
Sex, *n* (%)	
Male	716 (68.3)
Female	333 (31.7)
Race, *n* (%)	
White	756 (72.1)
Other	44 (4.2)
Not reported	249 (23.7)
Sarcomatoid features, *n* (%)	
No	855 (81.5)
Yes	90 (8.6)
Not reported	104 (9.9)
Primary tumor grade, *n* (%)	
1	54 (5.1)
2	461 (43.9)
3	362 (34.5)
4	163 (15.5)
NA	9 (0.9)
Pathologic T stage, *n* (%)	
T1	521 (49.7)
T2	95 (9.1)
T3	393 (37.5)
T4	17 (1.6)
NA	23 (2.2)
Pathologic node stage, *n* (%)	
N0	577 (55)
N1/N2	51 (4.9)
Nx	397 (37.8)
NA	24 (2.3)
Metastatic stage, *n* (%)	
M0	739 (70.4)
M1	166 (15.8)
Mx	124 (11.8)
NA	20 (2.0)
AJCC pathologic stage, *n* (%)	
I	507 (48.3)

Characteristic	Composite cohort, *n* = 1049
II	77 (7.3)
III	276 (26.3)
IV	189 (18.0)
Genes	
VHL, % mutated	63.49
WT, *n*	383
Mutated, *n*	666
PBRM1, % mutated	37.18
WT, *n*	659
Mutated, *n*	390
SETD2, % mutated	14.20
WT, *n*	900
Mutated, *n*	149
BAP1, % mutated	11.06
WT, *n*	933
Mutated, *n*	116
KDM5C, % mutated	7.43
WT, *n*	847
Mutated, *n*	68
NA, *n*	134
TP53, % mutated	3.44
WT, *n*	757
Mutated, *n*	27
NA, *n*	265
MTOR, % mutated	6.51
WT, *n*	733
Mutated, *n*	51
NA, *n*	265
PTEN, % mutated	4.08
WT, *n*	752
Mutated, *n*	32
NA, *n*	265
TSC1, % mutated	1.66
WT, *n*	771
Mutated, *n*	13
NA, *n*	265
TSC2, % mutated	1.15
WT, *n*	775
Mutated, *n*	9
NA, *n*	265
NF2, % mutated	1.28
WT, *n*	774

Characteristic	Composite cohort, *n* = 1049
Mutated, *n*	10
NA, *n*	265
PIK3CA, % mutated	2.81
WT, *n*	762
Mutated, *n*	22
NA, *n*	265
KEAP1, % mutated	1.40
WT, *n*	773
Mutated, *n*	11
NA, *n*	265
TET2, % mutated	2.55
WT, *n*	764
Mutated, *n*	20
NA, *n*	265

AJCC = American Joint Committee on Cancer; BMI = body mass index; NA = not available; SSIGN = Stage, Size, Grade, and Necrosis; WT = wild type.

Table 2

Multivariable competing risks regression for cancer-specific survival with the Stage, Size, Grade, and Necrosis set of adjustment variables

	HR (95% CI)	*q* value
BAP1	1.29 (0.85–1.95)	0.230
TP53	2.23 (1.27–3.92)	0.005
Age	1.02 (1–1.04)	0.022
Sex*	1.19 (0.81–1.74)	0.390
SSIGN score	1.42 (1.35–1.49)	<0.001

CI = confidence interval; HR = hazard ratio; SSIGN = Stage, Size, Grade, and Necrosis. Competing risks regression adjusted for everything listed in the table as well as cohort. There were 140 events among 554 patients included in multivariable analysis.

*Reference sex is female.

Table 3

Multivariable competing risks regression for cancer-specific survival adjusted for the base set of variables and sarcomatoid features

	HR (95% CI)	*q* value
BAP1	1.87 (1.30–2.68)	0.001
TP53	2.03 (1.05–3.90)	0.034
Age	1.02 (1–1.04)	0.001
Sex *	1.14 (1.01–2.05)	0.390
Stage †	7.99 (5.62–11.36)	<0.001
Sarcomatoid features	2.80 (1.70–4.59)	<0.001

CI = confidence interval; HR = hazard ratio.

* Reference sex is female.

† Stage IV versus stages I/II/III.

Competing risks regression adjusted for everything listed in the table as well as cohort. There are 160 events among 680 patients included in multivariable analysis.

Table 4

Multivariable competing risks regression for recurrence-free survival adjusted for the Stage, Size, Grade, and Necrosis set of variables

	HR (95% CI)	*q* value
SETD2	1.67 (1.01–2.77)	0.047
KDM5C	1.48 (0.82–2.68)	0.200
Age	1.01 (0.99–1.03)	0.280
Sex *	1.18 (0.74–1.87)	0.480
SSIGN score †	1.44 (1.33–1.55)	<0.001

CI = confidence interval; HR = hazard ratio; SSIGN = Stage, Size, Grade, and Necrosis.

Competing risks regression adjusted for SSIGN set of variables as well as site. There are 114 events among 539 patients included in multivariable analysis.

* Reference sex is female.

† Patients with stage IV disease were not eligible for analysis of recurrence.