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ORIGINAL ARTICLE

Prognostic significance of BAP1 expression in high-grade upper tract urothelial carcinoma: a multi-institutional study

Ahmet M. Aydin[1,2] · Nirmish Singla[1] · Vandana Panwar[3] · Solomon L. Woldu[1] · Yuval Freifeld[1] · Christopher G. Wood[4] · Jose A. Karam[4] · Alon Z. Weizer[5] · Jay D. Raman[6] · Mesut Remzi[7] · Nathalie Rioux-Leclercq[8] · Andrea Haitel[9] · Marco Roscigno[10] · Christian Bolenz[11] · Karim Bensalah[12] · Mary E. Westerman[1,13] · Arthur I. Sagalowsky[1] · Shahrokh F. Shariat[7] · Yair Lotan[1] · Aditya Bagrodia[1] · Payal Kapur[3] · Vitaly Margulis[1] · Laura-Maria Krabbe[1,14]

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Abstract

Purpose To evaluate the prognostic value of BRCA1-associated protein-1 (BAP1) expression in upper tract urothelial carcinoma (UTUC), as *BAP1* mutations have been associated with prognostic implications in urologic and non-urologic malignancies.

Methods We reviewed a multi-institutional cohort of patients who underwent radical nephroureterectomy (RNU) for high-grade UTUC from 1990–2008. Immunohistochemistry (IHC) for BAP1 was performed on tissue microarrays. Staining intensity was graded from 0–3, with BAP1 loss defined as an average intensity of < 1. Clinicopathologic characteristics and oncologic outcomes [recurrencefree (RFS), cancer-specific (CSS), and overall survival (OS)] were stratified by BAP1 status. The prognostic role of BAP1 was assessed using Kaplan–Meier (KM) and Cox regression analysis. Significance was defined as $p < 0.05$.

Results 348 patients were included for analysis and 173 (49.7%) showed BAP1 loss. Median follow-up was 36.0 months. BAP1 loss was associated with papillary architecture and absence of tumor necrosis or CIS. On univariable analysis, BAP1 loss was associated with improved RFS (HR 0.60, $p = 0.013$) and CSS (HR 0.55, $p = 0.007$), although significance was lost on multivariable analysis (HR 0.71, $p = 0.115$ and HR 0.65, $p = 0.071$; respectively) after adjusting for other significant parameters. BAP1 expression was not significantly associated with OS.

Conclusions BAP1 loss was associated with favorable pathologic features and better oncologic outcomes in univariate but not multivariate analysis in patients with high-grade UTUC. In contrast to renal cell carcinoma, loss of BAP1 expression appears to confer a better prognosis in high-grade UTUC. The role of the *BAP1* pathway in UTUC pathogenesis remains to be further elucidated.

Keywords BAP1 protein · Human · Biomarkers · Carcinoma · Transitional cell · Prognosis · Urinary tract

Introduction

Upper tract urothelial carcinoma (UTUC) is an uncommon genitourinary malignancy, which accounts for approximately 5% of all urothelial carcinomas [1]. It concurrently occurs with bladder cancer in approximately 15% of cases and may also present as a hereditary extracolonic manifestation of Lynch Syndrome [2].

Radical nephroureterectomy (RNU) with bladder cuff excision is the standard treatment for localized UTUC. RNU is associated with durable local control and cancer-specific survivalhowever survival rates decrease dramatically in patients with advanced disease [3]. These patients

Ahmet M. Aydin and Nirmish Singla contributed equally.

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✉ Vitaly Margulis
vitaly.margulis@utsouthwestern.edu

Extended author information available on the last page of the article

might benefit from perioperative systemic therapyhowever selection criteria beyond classical pathological parameters are currently lacking [4, 5]. Several nomograms and tissue/blood-based biomarkers have been proposed, but none are recommended for routine clinical use, given retrospective design and small sample size of these studies [1]. Novel prognostic biomarkers with high accuracy are needed to stratify patients for personalized treatment and follow-up since cancer-specific survival (CSS) of patients with UTUC improved only marginally over the past three decades [6].

The BRCA1-associated protein-1 (BAP1) is a nuclear-localized, ubiquitin carboxy-terminal hydrolase, which is involved in the removal of ubiquitin from proteins and histone H2A [7]. The *BAP1* gene is located on chromosome 3p21.1 [7]. It acts as a tumor suppressor by binding to the wild-type BRCA1-RING finger domain [8]. Its deubiquitinating activity enhances growth control of BRCA1, which is an essential player in DNA damage repair [8]. *BRCA1* mutations are related to cellular accumulation of chromosomal abnormalities and tumorigenesis in many cancers [8]. BAP1 also interacts with HCF-1, a protein that binds to the histone-modifying complex. During cell division it enhances the progression through the G1-S checkpoint and can also induce cell death [9, 10].

The significance of BAP1 expression in prognosis and survival outcomes has been reported in several malignancies such as renal cell carcinoma and uveal melanoma, where loss of BAP1 confers worse as well as malignant mesothelioma where loss of BAP1 projects favorable prognosis [11–13]. Our current knowledge about the role of BAP1 expression in UTUC is very limited. However, the presence of DNA damage repair mutations (DDR) and microsatelliteinstability (MSI) is enriched in UTUC, providing an actionable target for systemic therapies [14]. In this multi-institutional study, we sought to evaluate the prognostic significance of BAP1 expression in high-grade UTUC.

Materials and methods

Patient selection and data collection

The previously described study population consisted of patients with high-grade UTUC treated with extirpative surgery retrieved from the Upper Tract Urothelial Carcinoma Collaboration [15]. Institutional review board (IRB) approval was obtained at each center. Clinicopathologic data and pathologic specimens for tissue microarray (TMA) construction were available for 753 patients treated with RNU between 1990 and 2008. For this study, patients with low-grade disease ($n=188$), history of neoadjuvant therapy ($n=20$), muscle-invasive bladder cancer ($n=31$) and UTUC at the uretero-enteric anastomosis ($n=7$) were excluded from the study as well as patients without enough tumor tissue on TMA ($n=159$).

Pathological evaluation and immunohistochemistry

Pathologic specimens were reviewed by experienced genitourinary pathologists, blinded to clinical outcomes. Tumors were staged according to the AJCC/UICC 2002 TNM classification and grading was assessed per the 1998 WHO/ISUP consensus classification [16, 17]. Tumor location, tumor architecture, presence of lymphovascular invasion (LVI), tumor necrosis, and concomitant carcinoma in situ (CIS) were evaluated in all specimens.

TMAs were created utilizing cores from each formalin-fixed, paraffin-embedded pathologic specimen. Immunohistochemical staining was performed for BAP1 utilizing a monoclonal mouse antibody (clone C-4, 1:50, Santa Cruz Biotechnology Inc., CA, USA). Slides were analyzed by manual read of 2 dedicated genitourinary pathologists. Appropriate positive and negative controls were used for each run of immunostains and checked for validation.

Nuclear staining intensity of BAP1 was evaluated Fig. 1 (supplemental) and positive nuclear staining in background stromal cells and intratumor lymphocytes served as internal positive controls. The intensity of staining was graded from 0 to 3 and BAP1 loss was defined as average staining intensity < 1.

Management and follow-up

Data accrual for sociodemographic, pathologic and follow-up data was closed before TMA construction. Median follow-up time was calculated utilizing patients who did not have a reported event of death until data accrual closure. Operative technique for RNU was not standardized across institutions. Use and extent of lymphadenectomy based on surgeon discretion. Patient follow-up generally occurred every 3 months in the first post-operative year, every 6 months in the second year, and once a year thereafter. Follow-up visits consisted of history and physical examination, laboratory measurements, urinary cytology, cystoscopic evaluation of the lower urinary tract, and cross-sectional imaging of the contralateral upper urinary tract. Recurrence was defined as locoregional recurrence in the renal fossa/pelvis depending on primary tumor location or systemic spread. Tumor findings within the bladder or the contrary upper urinary tract were coded as secondary malignancies. Cause of death was determined by chart review or death certificate.

Statistical

Patients were grouped as 'BAP1 positive' and 'BAP1 loss' following immunohistochemical evaluation. Clinicopathologic variables between the groups were compared using independent-sample Mann–Whitney *U* and Chi-square tests and oncologic outcomes [recurrencefree (RFS), cancer-specific (CSS), and overall survival (OS)] were compared using Kaplan–Meier (KM) method stratified by BAP1 status, analyzed with the log rank statistic. Independent prognosticators of oncologic outcomes (RFS, CSS, OS) were identified using univariable and multivariable Cox proportional hazard regression models. Harrell's C indices and the Net Reclassification Improvement (NRI) were calculated as described before [15]. All statistical were conducted using SPSS version 22.0 (IBM, Armonk, NY). *p* values are two-sided, with statistical significance defined as $p<0.05$. Post-hoc power analysis was performed using the generated hazard ratios (HR) for overall all-cause-mortality, at a significance level of 5% (two-sided) using PASS software [18, 19].

Results

Descriptive statistics are shown in Table 1. Median age was 70.0 at the time of surgery and median follow-up was 36.0 months for the patients alive at time of analysis. Lymph node dissection (LND) was performed in 24.7% of patients with a documented lymph node yield of one or more nodes (range 1–24).

Overall, 49.7% of patients showed BAP1 loss and were more likely to be male (53.2% vs. 40.6%). pT stage, pN stage, lymphovascular invasion and tumor focality were not associated with BAP1 status. BAP1 loss was associated with more favorable pathological features such as papillary tumor ($p=0.002$), absence of necrosis and lack of concomitant CIS ($p=0.013$ and $p=0.007$) (Table 1).

At the time of this analysis, recurrence of UTUC was noted in 27.9% of patients while death of UTUC and death of any cause were recorded in 24.7% and 38.5% of patients, respectively. Patients with BAP1 loss had improved RFS [144.4 months (95% CI 130.5–158.3) vs. 81.1 months (95% CI 70.0–92.2), $p=0.011$] and CSS [151.6 months (95% CI 138.2–165.0) vs. 103.2 months (95%CI 88.1–118.4), $p=0.006$] compared to those with positive BAP1 expression in KM and a trend towards significance was noted for OS [108.6 months (95%CI 92.0–125.2) vs. 83.8 months (95% CI 68.6–98.9), $p=0.058$] (Fig. 1).

On univariable analysis (UVA), BAP1 loss was associated with improved RFS (HR 0.60, $p=0.013$) and CSS (HR 0.55, $p=0.007$) (Table 2a). However the significance of BAP1 expression was lost for both RFS (HR 0.71, $p=0.115$) and CSS (HR 0.65, $p=0.071$) in multivariable analysis (MVA) that adjusted for the effects of clinicopathological parameters (Table 2b). There was no significant association between BAP1 status and OS in both UVA (HR 0.72, $p=0.60$) and MVA (HR 0.87, $p=0.467$).

Performance of the MVA models w slightly better with addition of BAP1 status. Harrell's C indices were 0.810 and 0.813 for RFS, 0.813 and 0.821 for CSS and 0.784 and 0.785 for OS in the models without and with BAP1 status, respectively. NRI was significant for RFS and CSS (0.05 each) but not for OS, with best reclassification in the event group. A sample of 348 patients with an overall death rate of 38.5% provided a statistical power (β) of 0.906 and 0.509 for HRs of 1.8 and 1.4, respectively.

Discussion

BAP1 is a tumor suppressor gene that plays a significant role in cell cycle regulation and apoptosis [9, 10]. Its mutation, resulting in BAP1 loss is thought to lead to cellular accumulation of chromosomal abnormalities and tumorigenesis in many cancers [8]. Among genitourinary cancers, prognostic significance of BAP1 has been best demonstrated in clear cell renal cell carcinomas (ccRCC), where loss of BAP1 expression is significantly associated with aggressive histopathological features and poor survival outcomes [12, 20]. Furthermore BAP1 mutant ccRCC, harboring a distinct tumor biology has been recently described [21].

In our study, approximately half of patients with high-grade UTUC were found to have BAP1 loss. BAP1 loss was more frequent in male patients although there was no survival difference between both sexes. Therefore BAP1 loss did not appear to be a mutation that selectively affected survival of male patients. Interestingly, BAP1 loss rather than its positivity was associated with favorable pathological features and lower probability of recurrence and cancer-specific mortality on univariate analysis. Our findings about the significance of BAP1 expression in UTUC were most consistent with the reported findings in malignant mesothelioma (MM). Farzin et al. reported that BAP1 loss in MM was associated with younger age at onset, epitheloid histology and improved median survival (16.1 vs. 6.3 months, $p<0.01$) [22]. Similarly, BAP1 loss seems to be associated with better survival outcomes in intrahepatic cholangiocarcinoma [23].

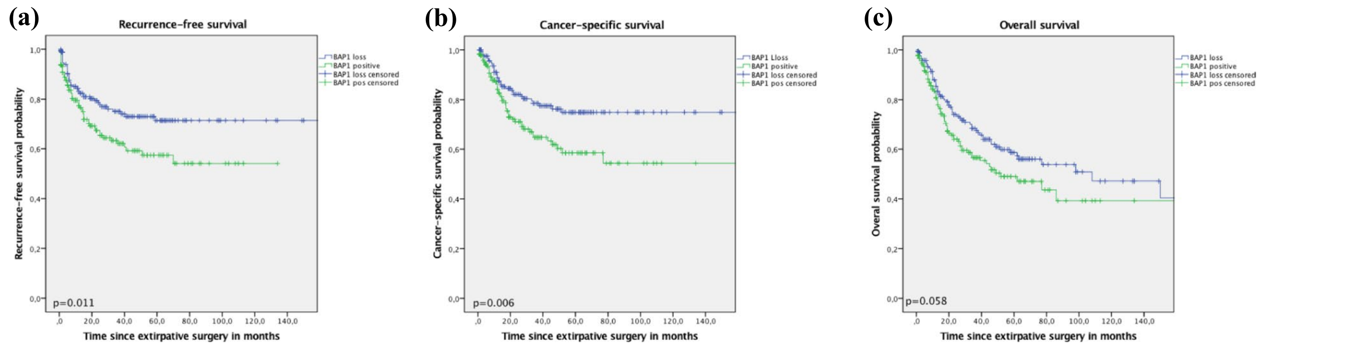
In our study, IHC analysis for BAP1 expression was a manually read semiquantative analysis and done via a validated scoring protocol for RCC, due to lack of previous reports for UTUC [20]. We did not perform confirmatory mutation analysis, but IHC was previously found to be sufficiently accurate for detection of BAP1 loss/inactivation [24]. Negative nuclear staining has been shown to correlate well with biallelic BAP1 genomic loss in various cancer types

Table 1 Association of BAP1 expression with clinical and pathological parameters in 348 patients treated with RNU for high-grade UTUC

Variables	Total	BAP1 expression status		*p* value
		Loss	Positive	
Number of patients, *n* (%)	348 (100)	173 (49.7)	175 (50.3)	–
Median age (Q1–3), years	70.0 (64.0–77.0)	69.0 (64.0–76.0)	70.3 (63.0–78.0)	0.388
Gender, *n* (%)				
Female	185 (53.2)	81 (46.8)	104 (59.4)	0.018
Male	163 (46.8)	92 (53.2)	71 (40.6)	
Location, *n* (%)				
Pelvicalyceal	267 (76.7)	143 (82.7)	124 (70.9)	0.009
Ureteral	81 (23.3)	30 (17.3)	51 (29.1)	
Focality, *n* (%)				
Unifocal	270 (77.6)	129 (74.6)	141 (80.6)	0.179
Multifocal	78 (22.4)	44 (25.4)	34 (19.4)	
Pathologic stage, *n* (%)				
pTis/pTa	31 (8.9)	12 (6.9)	19 (10.9)	0.386
pT1	103 (29.6)	56 (32.4)	47 (26.9)	
pT2	57 (16.4)	31 (17.9)	26 (14.9)	
pT3	129 (37.1)	63 (36.4)	66 (37.7)	
pT4	28 (8.0)	11 (6.4)	17 (9.7)	
pN stage, *n* (%)				
pN0	52 (14.9)	20 (11.6)	32 (18.3)	0.091
pNx	262 (75.3)	139 (80.3)	123 (70.3)	
pN+	34 (9.8)	14 (8.1)	20 (11.4)	
Organ confined, *n* (%)				
Yes (T stage ≤ 2, N0/x, M0)	185 (53.2)	96 (55.5)	89 (50.9)	0.386
No (T stage > 2 and/or N+)	163 (46.8)	77 (44.5)	86 (49.1)	
Lymphovascular invasion, *n* (%)				
Absent	250 (71.8)	129 (74.6)	121 (69.1)	0.261
Present	98 (28.2)	44 (25.4)	54 (30.9)	
Architecture, *n* (%)				
Papillary	286 (82.2)	153 (88.4)	133 (76.0)	0.002
Sessile	62 (17.8)	20 (11.6)	42 (24.0)	
Necrosis, *n* (%)				
Absent	286 (82.2)	151 (87.3)	135 (77.1)	0.013
Present	62 (17.8)	22 (12.7)	40 (22.9)	
Concomitant CIS				
Absent	275 (79.0)	147 (85.0)	128 (73.1)	0.007
Present	73 (21.0)	26 (15.0)	47 (26.9)	
Previous NMIBC				
Absent	251 (72.1)	133 (76.9)	118 (67.4)	0.875
Present	97 (27.9)	40 (23.1)	57 (32.6)	
Recurrence of UTUC				
No	251 (72.1)	133 (76.9)	118 (67.4)	0.011*
Yes	97 (27.9)	40 (23.1)	57 (32.6)	
Death of UTUC				
No	262 (75.3)	138 (79.8)	124 (70.9)	0.006*
Yes	86 (24.7)	35 (20.2)	51 (29.1)	
Death of any cause				
No	214 (61.5)	108 (62.4)	106 (60.6)	0.058*
Yes	134 (38.5)	65 (37.6)	69 (39.4)	

CIS carcinoma in situ, *NMIBC* non-invasive bladder carcinoma

*Log rank statistic



(a) Patients at risk

Time in months	0	12	24	48	96
BAP1 loss	173	123	95	59	17
BAP1 Positive	175	104	70	35	6
Cumulative events					
BAP1 loss	0	26	33	39	40
BAP1 Positive	0	35	48	55	57

(b) Patients at risk

Time in months	0	12	24	48	96
BAP1 loss	173	131	97	61	19
BAP1 Positive	175	166	76	38	7
Cumulative events					
BAP1 loss	0	15	26	32	33
BAP1 Positive	0	22	40	48	51

(c) Patients at risk

Time in months	0	12	24	48	96
BAP1 loss	173	131	97	61	19
BAP1 Positive	175	116	76	38	7
Cumulative events					
BAP1 loss	0	21	39	53	56
BAP1 Positive	0	28	52	64	69

Fig. 1 Recurrencefree (**a**), cancer-specific (**b**) and overall (**c**) survival probability stratified by BAP1 expression for 348 patients treated with radical nephroureterectomy for high-grade upper tract urothelial carcinoma

[24]. Pena-Llopis et al. reported that positive and negative predictive value of BAP1 IHC in ccRCC tumorgrafts were 100% and 98.6%, respectively [25].

Recently, multiple genomic characterization studies were performed for urothelial carcinoma, however mostly for UCB and not UTUC. They show rates of genomic BAP1 alterations of 7–15% for UCB [26, 27]. In UTUC, Sfakianos et al. analyzed somatic mutations and copy number alterations using next-generationsequencing (NGS) in 83 patients and reported BAP1 alterations in 4% of patients, although they did not analyze rare mutations and epigenetic changes [28]. Moss et al. performed whole-exome sequencing of DNA, RNA sequencing and protein analysis in 31 snap-frozen UTUC samples, but BAP1 did not make to list of genes that had > 10% alteration frequency in their cohort [29]. The frequency of BAP1 loss on the protein level detected by IHC in our study (49.7%) was notably higher than the frequency of BAP1 mutations/alterations reported in those studies by genomic sequencing. There are multiple theories to try to explain this observation. First, a NGS-based approach to detect mutations has many obstacles such as low DNA quantity from diagnostic biopsies, difficulty in processing of FFPE samples and tumor heterogeneity within samples which might cause reproducibility and generalizability issues [30].

Moreover, patients might have had more advanced disease with unfavorable features in our study (all were high grade, almost half were non-organ-confined). Our sample size is much larger than in the other studies of BAP1 in UTUC (348 vs. 31 and 83therefore the rate of mutations might have been underestimated in those studies [28, 29]. As DDR-gene alterations and MSI are enriched in UTUC, we might expect higher rates of alteration of associated genes as well [14]. Further, there no data of how expression of BAP1 on the protein level in UTUC is driven only by the necessity of a genomic mutation, or if dysregulation of other pathways might contribute to this discrepancy. , in this TMA, the tumor sections utilized were taken from the midst of the primary tumor (most aggressive and largest lesion), possibly influencing our findings as well.

Urothelial carcinomas show among highest numbers of mutations in solid cancers, so additional driver genes and their mutations may cause cooperative or antagonistic effects and may complement each other during carcinogenesis and progression of UTUC. For instance, an antagonistic effect of p53 might have altered the biological effects of BAP1 expression in our patients with advanced UTUC since p53 expression is associated with high grade/stage and its mutation is found in up to 30–60% of UTUCs [31]. Rare coincidence of loss of BAP1 and PBRM1, which is a tumor suppressor gene and shares the same gene locus, was associated with very poor prognosis in ccRCChowever we did not find any prognostic value of PBRM1 expression analysis by itself or in conjunction with BAP1 in our cohort (data not shown) [32]. *KDM6A* mutations can occur concomitantly with *BAP1* mutationshowever the implications of a combined loss are largely unknown [26].

Table 2 Univariable (a) and multivariable (b) Cox regression analyes for prognostication of disease recurrence, cancer-specific and overall mortality of 348 patients treated with radical nephroureterectomy for high-grade upper tract urothelial carcinoma

Variable	Recurrence-free survival			Cancer-specific survival			Overall survival		
	HR	95% CI	*p* value	HR	95% CI	*p* value	HR	95% CI	*p* value
(a) Univariable									
Age							1.04	1.02–1.06	< 0.001
Multifocality	1.72	1.07–2.78	0.026	1.88	1.12–3.18	0.018	1.83	1.21–2.76	0.004
pTstage	Overall		< 0.001	Overall		< 0.001	Overall		< 0.001
pTIS/pTa/pT1	–	–	–	–	–	–	–	–	–
pT2	2.46	1.09–5.55	0.03	2.69	1.06–6.82	0.037	1.5	0.80–2.82	0.21
pT3	4.64	2.30–9.34	< 0.001	6	2.70–13.3	< 0.001	3.04	1.81–5.12	< 0.001
pT4	21.5	9.00–51.4	< 0.001	24.4	8.96–66.6	< 0.001	10.5	4.88–22.5	< 0.001
pN-stage	Overall		0.003	Overall		0.128	Overall	Overall	0.196
pN0	–	–	–	–	–	–	–	–	–
pNx	0.89	0.49–1.61	0.697	1.1	0.55–2.20	0.78	1.05	0.61–1.79	0.868
pN+	2.18	1.07–4.42	0.031	1.96	0.86–4.45	0.11	1.69	0.84–3.39	0.141
Lymphovascular invasion	1.16	0.73–1.85	0.525	1.32	0.80–2.18	0.281	1.69	0.84–3.39	0.161
Sessile architecture	1.83	1.09–3.07	0.022	1.69	0.94–3.07	0.081	1.73	1.06–2.83	0.028
Necrosis	0.65	0.37–1.16	0.147	0.56	0.28–1.10	0.091	0.87	0.52–1.46	0.604
Concomitant CIS	1.25	0.74–2.12	0.399	0.89	0.48–1.66	0.894	0.91	0.55–1.52	0.729
BAP1 loss	0.71	0.46–1.09	0.115	0.65	0.41–2.04	0.071	0.87	0.60–1.26	0.467
(b) Multivariable									
Age	1	0.98–1.02	0.833	1.01	0.99–1.03	0.454	1.03	1.01–1.05	0.001
Female gender	0.87	0.58–1.30	0.869	0.84	0.55–1.29	0.425	0.98	0.70–1.39	0.928
Pelvicalyceal location	0.72	0.43–1.21	0.214	0.77	0.44–1.34	0.354	0.81	0.52–1.26	0.344
Multifocality	1.87	1.22–2.86	0.004	1.9	1.20–3.01	0.006	1.8	1.24–2.61	0.002
pTstage	Overall		< 0.001	Overall		< 0.001	Overall		< 0.001
pTIS/pTa/pT1	–	–	–	–	–	–	–	–	–
pT2	3.09	1.40–6.81	0.005	3.31	1.33–8.23	0.01	1.77	0.96–3.27	0.068
pT3	6.43	3.35–12.4	< 0.001	7.9	3.72–16.8	< 0.001	4.05	2.54–6.46	< 0.001
pT4	35.8	12.6–77.0	< 0.001	45.3	19.4–106	< 0.001	18.9	10.3–34.9	< 0.001
pN-stage	Overall		< 0.001	Overall		< 0.001	Overall		< 0.001
pN0	–	–	–	–	–	–	–	–	–
pNx	0.88	0.49–1.56	0.651	1.07	0.55–2.10	0.844	1.02	0.61–1.71	0.944
pN+	4.24	2.16–8.31	< 0.001	4.75	2.22–10.2	<0.001	3.13	1.66–5.90	< 0.001
Lymphovascular invasion	2.64	1.77–3.96	< 0.001	2.94	1.91–4.53	<0.001	2.66	1.88–3.78	< 0.001
Sessile architecture	3.19	2.07–4.94	< 0.001	3.13	1.98–4.97	<0.001	2.61	1.78–3.84	< 0.001
Necrosis	1.6	0.98–2.63	0.061	1.69	0.99–2.88	0.055	1.92	1.26–2.92	0.002
Concomitant CIS	1.77	1.13–2.76	0.012	1.44	0.87–2.37	0.159	1.4	0.93–2.11	0.104
Previous NMIBC	1.05	0.69–1.59	0.833	1	0.63–1.59	0.992	1.26	0.88–1.80	0.21
BAP1 loss	0.6	0.40–0.90	0.013	0.55	0.35–0.85	0.007	0.72	0.51–1.01	0.06

CI confidence interval, *CIS* carcinoma in situ, *HR* hazard ratio, *NMIBC* non-invasive bladder cancer

There are important limitations to our study mostly related to its retrospective design in a multi-institutional setting without central pathological review. However, all staining and interpretation performed by expert genitourinary pathologists (PK, VP), who were blinded to clinical outcomes and had special expertise for BAP1 expression in RCC [20]. Some of the shortcomings are related to the wide range of the study period as well as the retrospective nature of the study: e.g. the technique of RNU as well as the performance of lymph node dissection was at the discretion of the surgeon. Forty-five percent of the whole patient cohort had non-organ confined diseasehowever the rate of LND was 25% in this study. Although low utilization of LND might have hampered exact pathological tumor staging, the indication is not always clear upfront due to limitations in detection of invasive UTUC at the time of preoperative staging. Due to the multi-institutional and multi-national setting of the study with many surgeons included, there was no exact information about the technique of bladder cuff excision. Moreover use of adjuvant chemotherapy (AC) was not standardized across the institutions. AC was used in only around 30 patientsnevertheless it might have been selectively offered to the patients with worst prognosis, best performance status and most importantly normal renal function. However, the use of AC did not alter our findings (data not shown).

Median follow-up of 36 months could be considered low for such cohort. However, most UTUC patients who experience a relevant recurrence and subsequent death do so within the first years after RNU [33]. Contrary to observed favorable association between BAP1 loss and RFS/CSS, there was no such association to OS in our study. Non-cancer-specific causes of mortality such as co-morbidities and socioeconomic factors might have been quite diverse due to the multi-institutional/national design and might have obscured the association between BAP1 loss and OS. The confidence intervals of HRs were relatively large, which might be due to high variability or sample size of the study. However, to evaluate the validity of study findings, we performed a post-hoc power analysis and demonstrated that the study power reached acceptable levels. Nevertheless, such power analysis should be interpreted cautiously since it was conducted on observed insignificant results and performed on retrospective cohorts reliant on patients available for evaluation. Moreover, rather than low study power, there might actually be a real difference between groups less than the hypothesized effect size or no difference at all.

, this is the description of one marker only, and a combination of markers with analysis of mutations and correlated protein expressions might have enhanced the performance of the model. However, BAP1 loss is a recognized event in carcinogenesis utilized as an individual prognostic marker in multiple tumor entitiestherefore, its evaluation for UTUC seems reasonable.

Conclusions

This is the first study to demonstrate that BAP1 loss was associated with favorable clinicopathological features in patients with high-grade UTUC treated with extirpative surgery. Although BAP1 loss was associated with better recurrencefree and cancer-specific survival in univariable analysis, it did not emerge as an independent prognostic factor after adjustment for other clinicopathologic characteristics. Interestingly, our findings seem to parallel those of mesothelioma and intrahepatic cholangiocarcinoma, in which BAP1 loss confers a better prognosis, contrary to its association with poor disease outcomes in several malignancies.

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Availability of materials and data All data generated or analyzed during this study are included in this published article.

Compliance with ethical standards

Conflict of interest We declare that there are no potential competing interests in this research.

Ethical standards This study was conducted according to the Declaration of Helsinki and institutional review board approval was obtained at each center participating in this research. An institutional IRB was granted through a local ethics committee at each location.

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Affiliations

Ahmet M. Aydin[1,2] · Nirmish Singla[1] · Vandana Panwar[3] · Solomon L. Woldu[1] · Yuval Freifeld[1] · Christopher G. Wood[4] · Jose A. Karam[4] · Alon Z. Weizer[5] · Jay D. Raman[6] · Mesut Remzi[7] · Nathalie Rioux-Leclercq[8] · Andrea Haitel[9] · Marco Roscigno[10] · Christian Bolenz[11] · Karim Bensalah[12] · Mary E. Westerman[1,13] · Arthur I. Sagalowsky[1] · Shahrokh F. Shariat[7] · Yair Lotan[1] · Aditya Bagrodia[1] · Payal Kapur[3] · Vitaly Margulis[1] · Laura-Maria Krabbe[1,14]

[1] Department of Urology, University of Texas Southwestern Medical Center, 5323 Harry Hines Blvd., J8.130, Dallas, TX 75390-9110, USA

[2] Department of Urology, Hacettepe University, Ankara, Turkey

[3] Department of Pathology, University of Texas Southwestern Medical Center, Dallas, TX, USA

[4] Department of Urology, MD Anderson Cancer Center, Houston, TX, USA

[5] Department of Urology, University of Michigan Cancer Center, Ann Arbor, MI, USA

[6] Division of Urology, Penn State Milton S. Hershey Medical Center, Hershey, PA, USA

[7] Department of Urology, Medical University of Vienna, Vienna, Austria

[8] Department of Pathology, Centre Hospitalier Universitaire de Rennes, Rennes, France

[9] Department of Pathology, Medical University Vienna, Vienna, Austria

[10] Department of Urology, Ospedali Riuniti of Bergamo, Bergamo, Italy

[11] Department of Urology, University of Ulm, Ulm, Germany

[12] Department of Urology, Centre Hospitalier Universitaire de Rennes, Rennes, France

[13] Department of Urology, Mayo Clinic, Rochester, MN, USA

[14] Department of Urology, University of Muenster Medical Center, Muenster, Germany