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Makrakis, Dimitrios; Talukder, Rafee; Lin, Genevieve Ihsiu; Diamantopoulos, Leonidas N.; Dawsey, Scott; Gupta, Shilpa; Carril-Ajuria, Lucia; Castellano, Daniel; de Kouchkovsky, Ivan; Koshkin, Vadim S.; ...

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Dimitrios Makrakis MD[1]*, Rafee Talukder MD[1]*, Genevieve Ihsiu Lin MS[2], Leonidas N. Diamantopoulos MD[3], Scott Dawsey MD[4], Shilpa Gupta MD[4], Lucia Carril-Ajuria MD[5], Daniel Castellano MD[5], Ivan de Kouchkovsky MD[6], Vadim S. Koshkin MD[6], Joseph J. Park MD[7], Ajjai Alva MD[7], Mehmet A. Bilen MD[8], Tyler F. Stewart MD[9], Rana R. McKay MD[9], Nishita Tripathi MD[10], Neeraj Agarwal MD[10], Naomi Vather-Wu MD[11], Yousef Zakharia MD[12], Rafael Morales-Barrera MD[13], Michael E. Devitt MD[14], Alessio Cortellini MD[15], Fulgenzi Claudia Angela Maria MD[15], David J. Pinato MD, PhD[15], Ariel Nelson MD[16], Christopher J. Hoimes DO[17,18], Kavita Gupta MD[19], Benjamin A. Gartrell MD[19], Alex Sankin MD[19], Abhishek Tripathi MD[20], Roubini Zakopoulou MD, PhD[21], Aristotelis Bamias MD, PhD[22], Jure Murgic MD PhD[23], Ana Fröbe MD, PhD[23,24], Alejo Rodriguez-Vida MD, PhD[25], Alexandra Drakaki MD[26], Sandy Liu MD[26], Eric Lu, MD[26], Vivek Kumar MD[27], Giuseppe Di Lorenzo MD[28], Monika Joshi MD[29], Pedro Isaacsson-Velho MD[30,31], Lucia Alonso Buznego MD[32], Ignacio Duran MD[32], Marcus Moses MS[33], Albert Jang MD[33], Pedro Barata MD, MSc[33], Guru Sonpavde MD[34], Evan Y. Yu MD[1,35], Robert Bruce Montgomery, MD[1,35], Petros Grivas MD, PhD[1,35,]**, Ali Raza Khaki MD, MS[36]**

*: these authors contributed equally to this work
**: these authors contributed equally to this work

[1]*Division of Medical Oncology, Department of Medicine, University of Washington, Seattle, WA, USA.*
[2]*Department of Epidemiology, University of Washington, Seattle, WA, USA*
[3]*Department of Medicine, University of Pittsburgh, Pittsburgh, PA, USA*
[4]*Department of Hematology and Oncology, Cleveland Clinic Taussig Cancer Institute, Cleveland, Ohio, USA*
[5]*Department of Medical Oncology, Hospital Universitario 12 de Octubre, Madrid, Spain.*
[6]*Division of Oncology, Department of Medicine, University of California, San Francisco, San Francisco, CA, USA.*
[7]*Division of Oncology, Department of Medicine, University of Michigan, Ann Arbor, MI.*
[8]*Winship Cancer Institute of Emory University, Atlanta, GA.*
[9]*Division of Hematology/Oncology, Department of Medicine, University of California San Diego, La Jolla, CA.*
[10]*Division of Oncology, Department of Medicine, University of Utah, Salt Lake City, UT.*
[11]*Department of Medicine, University of Iowa, Iowa City, IA.*
[12]*Division of Oncology, Department of Medicine, University of Iowa, Iowa City, IA.*
[13]*Vall d'Hebron Institute of Oncology, Vall d' Hebron University Hospital, Universitat Autònoma de Barcelona, Barcelona, Spain.*
[14]*Division of Hematology/Oncology, Department of Medicine, University of Virginia School of Medicine, Charlottesville, VA.*
[15]*Department of Surgery and Cancer, Imperial College London, London, UK.*
[16]*Division of Hematology and Oncology, Department of Medicine, Medical College of Wisconsin, Milwaukee, WI*
[17]*Division of Medical Oncology, Seidman Cancer Center at Case Comprehensive Cancer Center, Cleveland, OH.*
[18]*Division of Medical Oncology, Duke University, Durham, NC.*
[19]*Departments of Medical Oncology and Urology, Montefiore Medical Center, Bronx, NY.*
[20]*Stephenson Cancer Center, University of Oklahoma Health Sciences Center, Oklahoma City, OK.*
[21]*Department of Clinical Therapeutics, Alexandra General Hospital, National and Kapodistrian*

University of Athens, School of Medicine, Athens, Greece.

[22]2nd Propaedeutic Dept of Internal Medicine, ATTIKON University Hospital, National and Kapodistrian University of Athens, School of Medicine, Athens, Greece.

[23]Department of Oncology and Nuclear Medicine, University Hospital Center Sestre Milosrdnice, Zagreb

[24]School of Dental Medicine, Zagreb, Croatia.

[25]Medical Oncology Department, Hospital del Mar Research Institute, Barcelona, Spain.

[26]Division of Hematology/Oncology, David Geffen School of Medicine at UCLA, Los Angeles, CA.

[27]Department of Medicine, Brigham and Women's Hospital, Harvard Medical School, Boston, MA.

[28]Oncology University of Molise and Medical Oncology ASL, Salerno, Italy.

[29]Division of Hematology/Oncology, Department of Medicine, Penn State Cancer Institute, Hershey, PA.

[30]Department of Oncology, Sidney Kimmel Comprehensive Cancer Center, Baltimore, MD

[31] Division of Oncology, Hospital Moinhos de Vento, Porto Alegre, Brazil.

[32]Hospital Universitario Marques de Valdecilla. IDIVAL. Santander, Spain.

[33]Deming Department of Medicine, Section of Hematology/Oncology, Tulane University, New Orleans, LA.

[34]Genitourinary Oncology Program, Dana-Farber Cancer Institute, Harvard Medical School, Boston, MA.

[35]Clinical Research Division, Fred Hutchinson Cancer Center, Seattle, WA.

[36]Division of Oncology, Department of Medicine, Stanford University, Palo Alto, CA.

Corresponding authors:

Ali Raza Khaki, M.D., M.S.
Clinical Assistant Professor, Division of Oncology, Stanford University
875 Blake Wilbur Dr., Stanford, CA 94305
Email: alikhaki@stanford.edu   Phone: 650-736-0519

Petros Grivas, M.D., Ph.D.
Associate Professor, Division of Medical Oncology, Dept. of Medicine, University of Washington School of Medicine; Clinical Research Division, Fred Hutchinson Cancer Center
1144 Eastlake Ave E, Mailstop: LG-465, Seattle, WA 98109, USA
Email: pgrivas@uw.edu   Phone: 206-606-1943

DECLARATION OF INTERESTS (Conflicts of Interest)

Dimitrios Makrakis has no COI to declare

Rafee Talukder has no COI to declare

Genevieve Ihsiu Lin has no COI to declare

Michael E. Devitt has no COI to declare.

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Micro-abstract:

Immune checkpoint inhibitors are a well-established treatment option for advanced urothelial carcinoma, and biomarkers of response are needed for better patient selection. We show that metastatic disease confined to lymph nodes is associated with better outcomes, while metastases to liver, bone or both are associated with poor outcomes with immune checkpoint inhibitor therapy. Results are hypothesis-generating but relevant to practice.

Abstract

Background: Sites of metastasis have prognostic significance in advanced urothelial carcinoma (aUC), but more information is needed regarding outcomes based on metastatic sites in patients treated with immune checkpoint inhibitors (ICI). We hypothesized that presence of liver/bone metastases would be associated with worse outcomes with ICI.

Methods: We identified a retrospective cohort of patients with aUC across 26 institutions, collecting demographics, clinicopathological, treatment, and outcomes information. Outcomes were compared with logistic (observed response rate; ORR) and Cox (progression-free survival; PFS, overall survival; OS) regression between patients with/without metastasis beyond lymph nodes (LN) and those with/without bone/liver/lung metastasis. Analysis was stratified by 1st or 2^{nd+} line.

Results: We identified 917 ICI-treated patients: in the 1st line, bone/liver metastases were associated with shorter PFS (HR: 1.65 and 2.54), OS (HR: 1.60 and 2.35, respectively) and lower ORR (OR: 0.48 and 0.31). In the 2^{nd+} line, bone/liver metastases were associated with shorter PFS (HR: 1.71 and 1.62), OS (HR: 1.76 and 1.56) and, for bone-only metastases, lower ORR (OR: 0.29). In the 1st line, LN-confined metastasis was associated with longer PFS (HR: 0.53), OS (HR:0.49) and higher ORR (OR: 2.97). In the 2^{nd+} line, LN-confined metastasis was associated with longer PFS (HR: 0.47), OS (HR: 0.54), and higher ORR (OR: 2.79); all associations were significant.

Conclusion: Bone and/or liver metastases were associated with worse, while LN-confined metastases were associated with better outcomes in patients with aUC receiving ICI. These findings in a large population treated outside clinical trials corroborate data from trial subset analyses.

Keywords: bladder cancer, urothelial carcinoma, immune checkpoint inhibitors, outcomes, metastasis

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Introduction

Locally advanced/unresectable or metastatic (herein described as 'advanced') urothelial carcinoma (aUC) can cause significant morbidity and mortality. Recently there have been major advances in the management of aUC with the use of immune checkpoint inhibitors (ICI) in the frontline, switch maintenance and salvage settings[1,2], as well as antibody-drug conjugates (enfortumab-vedotin, sacituzumab-govitecan) and the FGFR inhibitor erdafitinib in the salvage setting[3–5]. However, the observed response rates (ORR) and progression-free survival (PFS) achieved with ICI remain modest across treatment settings, while there is a considerable risk of immune related adverse events (IRAEs)[1]. This status emphasizes the need for the discovery and validation of accurate both prognostic and predictive biomarkers to help select patients more likely to benefit and reduce (or delay) ICI exposure to those who are unlikely to benefit. While predictive biomarkers have higher value in patient selection for particular therapies and optimal sequencing of agents in aUC, prognostic biomarkers can still help inform discussions about prognostic estimates, as well as in clinical trial design, stratification, and interpretation of results.

There have been several proposed clinical and molecular putative biomarkers associated with survival with ICI in aUC[6–11]. Several clinical factors, such as performance status, sites of metastasis (e.g. liver) albumin and hemoglobin levels have been shown to hold prognostic value for patients receiving ICI in the first (1st) line setting[10]. Moreover, data from clinical trials and retrospective studies suggest that patients with liver and bone metastasis who receive ICI have overall poor outcomes[12–14]. A retrospective study also has suggested that bone metastasis at time of aUC diagnosis is associated with poor outcomes with ICI.[15]

However, questions remain regarding the response and outcomes with ICI based on specific sites of metastasis and line of therapy, when adjusting, as possible, for other known prognostic factors. Additionally, data on the association between lung metastasis and response or outcomes with ICIs in aUC remains scarce at this point. Presence of liver, bone or lung metastases could potentially be associated with immunosuppression and poor response and outcomes with ICI. To further investigate the association between metastatic sites and outcomes with ICI, we evaluated response and survival with ICI in patients with aUC according to metastatic site. We hypothesized that response and survival with ICI would be inferior in patients with metastases not confined to lymph nodes (LNs), especially those with liver and/or bone metastases, as opposed to patients with LN-confined metastatic disease. We also hypothesized that there would be a significant interaction between the co-presence of liver and bone metastases regarding outcomes with ICI.

Patients & methods

Study design, patients and data collection

In concordance with the Declaration of Helsinki and after obtaining institutional review board approval, we performed a retrospective cohort study, using a cohort of patients from 25 institutions across the United States and Europe.[10,16–18] Consecutive patients at each institution were identified using a combination of provider-driven and electronic health record search algorithms.

Patients were included for analysis if they were treated with ICI monotherapy for aUC. We excluded patients who received ICI for a different cancer type or in a combination regimen or on a clinical trial, had pure non-urothelial histology, received more than one line of ICI therapy, or the sites of metastasis were unknown (**Figure 1-Consort diagram**). In addition, patients were excluded from the efficacy analysis, e.g. ORR, PFS, overall survival (OS) if the information regarding the endpoint was missing.

For data collection and storage, we used web-based, secure and standardized REDCap capture tools hosted at the Institute of Translational Sciences at the University of Washington[19,20]. Data collected included demographics, stage, histology type, prior therapies for earlier UC settings, laboratory values, sites of metastatic disease, laboratory parameters and clinical endpoints (e.g. response, progression, death). For metastatic sites, we collected information about metastasis to the following sites: lymph node, soft tissue, lung, liver, bone, adrenal gland, CNS, peritoneum, intestine. Information on metastases on other miscellaneous sites outside of the categories above, was also collected.

Pathology and radiology data were assessed from notes in the electronic health record based on comprehensive investigator review; no centralized review of images or pathology was feasible. All patients underwent imaging at the discretion of treating provider. Evaluation of response and progression were determined by the investigator review based on the best available information in clinic notes and radiographic study reports without formal central radiology review based on strict RECIST 1.1 criteria.

Statistical analysis

Baseline characteristics were summarized using descriptive statistics and compared via chi-square and Student's t-tests, for categorical and continuous variables, respectively. ORR was calculated as the number of patients with complete or partial response divided by the total number of patients. OS was defined as time from ICI initiation to date of death from any cause, and PFS as time from ICI initiation until date of radiographic or clinical progression or death from any cause. Patients who did not experience death or progression were censored at the date of last follow-up.

All outcomes (ORR, PFS, OS) were analyzed separately for patients treated with ICI in the first line (1st) and the subsequent setting (second line and beyond; 2^{nd+}). No patient with switch maintenance ICI was included. For our analysis, we created three separate models: in model A we compared patients with vs without LN confined metastasis; those with soft tissue recurrent/metastatic disease were classified as non-LN confined; in model B, we compared patients with no bone or liver metastasis with those having bone-only, liver-only, or both. We also tested the interaction between bone and liver metastases in this model, by modeling both bone and liver metastases as separate binary variables and including a multiplicative interaction term in the model. Finally, in model C, we compared patients with no bone, liver or lung metastasis with those having any combination of those three sites, such as liver-only, bone-only or lung-only, bone and liver without lung, liver and lung without bone, bone and lung without liver, or all three sites together. We also tested for any interaction between these sites (bone-liver, bone-lung, liver-lung, and bone-liver-lung).

We used univariable and multivariable logistic regression to estimate the odds ratios (OR) and confidence intervals (CI) for ORR dependent on exposure. For OS and PFS, we estimated median survival time using the Kaplan-Meier method. Hazard ratios (HR) dependent on exposure were calculated using univariable and multivariable Cox Regression. For multivariable modeling, we *a priori* identified factors hypothesized to possibly confound the relationship between metastatic site and efficacy endpoints (ORR, PFS, OS). We tested each factor independently in a model, retaining any variable for the final model that changed the outcome of interest (OR or HR) by >10%. The factors that were tested were age (as a continuous variable), sex (male, female), white race (yes, no), smoking history (yes, no), history of cystectomy [or (nephro)ureterectomy for upper tract UC] (yes, no), histology (pure urothelial carcinoma vs mixed), presence of upper tract urothelial carcinoma (yes, no) and ECOG PS (0-4, modeled as discrete categories) along with hemoglobin, albumin, and Neutrophil to Lymphocyte Ratio (NLR). Hemoglobin, albumin and NLR were all modeled as continuous variables. The alpha level was set at 0.05 for all analyses, which were performed with Stata IC 16.0 (Stata LLC, College Station, Texas).

Results

Patient characteristics

We analyzed data from 1,283 patients with aUC treated with ICIs across 26 institutions between 2013-2021. After exclusions, we identified 917 patients that were treated with ICI monotherapy for aUC. Ultimately, 886 were included in the ORR, 898 patients were included in the PFS and 876 in the OS analysis, respectively. Baseline characteristics of patients are shown on Table 1, stratified by line of therapy (1st and 2nd+). Among 504 patients treated with 1st line and 413 patients with 2nd+ line ICIs, pure urothelial histology was present in 69% and 74% of cases, respectively. At ICI initiation, LN-metastatic disease was present in 72% and 79% for 1st and 2nd+ line, respectively, while the prevalence of liver metastasis was 17% and 25% for 1st and 2nd+ line, of bone metastasis was 22% and 32%, and of lung metastasis was 32% and 39% respectively. (**Table 1**) Patient characteristics by site of metastasis (LN and bone, lung or liver) and line of therapy (1st or 2nd+) can be found in supplementary tables. (**Table S1-S4**).

Observed Response Rate

A total of 886 patients were included in the ORR analysis; of those, 479 received ICI in the 1st line and 407 in the 2nd+ line. In the model A ("LN model") analysis of both the 1st and 2nd+ lines, patients with LN-only metastasis had a significantly higher ORR compared to those with disease spread beyond LNs. (**Table 2**).

In the model B ("bone-liver model") analysis, the presence of bone metastases, liver metastases or both were each significantly associated with lower ORR vs those without bone or liver metastases in the 1st line setting; among patients treated with ICI as 2nd+ line, only bone metastases were associated with a significantly lower ORR (**Table 3**). However, in our third model (Model C) only liver metastases retained a significant association with lower ORR in the 1st line. Similarly, in the 2nd+ line, only bone and bone & lung metastases showed a significant association with lower ORR. Lung-only metastases were not significantly associated with lower ORR (**Table 4**). We did not identify a significant interaction between the presence of bone, liver or lung metastases in either 1st or 2nd+ line setting.

Progression-Free Survival

A total of 898 patients were included in the PFS analysis; of those, 495 were treated with ICI in the 1st line and 403 in the 2nd+ line. In both treatment settings, the presence of LN-confined metastasis was significantly associated with longer PFS compared to non-LN confined metastasis (**Table 2**; **Figure 2**).

In the model B (bone-liver) analysis, the presence of bone metastasis, liver metastasis or both were associated with shorter PFS compared to neither bone or liver, in both 1st line and 2nd+ line settings (**Table 3**). In Model C, liver-only metastasis, as well as co-presence of liver and bone, liver and lung, as well as liver, lung & bone metastases were associated with significantly shorter PFS in the 1st line setting. In the 2nd+ line, all those single sites (liver, lung, bone) of metastases as well as their combinations (liver-bone, liver-lung, bone-lung, and liver-bone-lung) were significantly associated with shorter PFS (**Table 4**). No significant interaction was detected between bone, liver or lung metastases for either 1st or 2nd+ line.

Overall Survival

A total of 876 patients were included in the OS analysis: 485 received 1st line ICI and 391 2nd+ line ICI. In the model A (LN model) analysis, patients treated with 1st line or 2nd+ ICIs with LN-confined metastasis had significantly longer OS compared to those without LN-confined metastasis (**Table 2**; **Figure 3**).

In the model B (bone-liver) analysis, patients with bone metastases, liver metastases or both had shorter OS compared to patients without either bone or liver metastases in both 1st line and 2nd+ line settings (**Table 3; Figure 3**). In the model C, liver-only metastases, along with bone and liver, lung and liver, as well as co-occurrence of bone, liver and lung metastases were significantly associated with shorter OS in the 1st line setting. In the 2nd+ line, bone-only metastases along with all combinations (bone and lung, bone and liver, liver and lung, as well as bone-liver-lung) were significantly associated with shorter OS (**Table 4**). Like ORR and PFS analyses, no significant interaction between liver and bone lesions was detected in either the 1st or the 2nd+ line.

Discussion

In this retrospective cohort of 917 patients with aUC treated with ICI outside clinical trials, we demonstrated that sites of metastases were associated with different outcomes on ICI therapy. Specifically, patients with aUC confined to LNs had significantly higher ORR, longer PFS and OS compared to patients without LN-confined disease, when treated with ICI as either 1st or 2nd+ line of therapy. Moreover, bone and liver metastases were associated with significantly lower ORR, shorter PFS and OS in both therapy settings. We did not identify a significant interaction between metastatic sites regarding outcomes with ICI therapy.

Previous studies have reported the association between specific sites of metastases and treatment outcomes in aUC. In a widely used prognostic model, Bajorin et al. identified the presence of visceral metastasis and poor ECOG PS to be associated with shorter OS in patients treated with 1st line platinum-based chemotherapy.[8] Similarly, the prognostic model by Bellmunt et al. in patients with platinum-refractory aUC treated with vinflunine chemotherapy identified liver metastasis, along with ECOG PS greater than 0 and Hgb<10 g/dL, as clinical factors associated with shorter OS. More recently, prognostic models developed in patients treated with ICI have also identified metastatic sites to have prognostic significance. A risk score developed by Khaki et al.[10] using an earlier data-lock of this cohort and assessing prognostic factors for 1st line ICI treatment in aUC, showed that liver metastases were associated with worse prognosis. For those treated with ICI in the 2nd+ line, a 5-factor prognostic model for response to ICI also identified liver metastasis as associated with significantly shorter OS[9], while another prognostic model by Nassar et al[11]. identified visceral metastasis as a poor prognostic feature.

While multiple prior prognostic models have specifically identified liver metastasis as a poor prognostic factor, an important additional finding in our study is the similarly poor prognostic outcomes with bone metastasis. Bone metastasis has been recently identified as a risk factor for worse outcomes in aUC. Nelson et al. reported a significant association between the presence of early bone metastases (defined as present at the time of metastatic diagnosis) and shorter OS for patients receiving either chemotherapy or ICI for aUC. In a separate analysis comparing the effect of bone metastasis to treatment with ICI vs chemotherapy, no significant difference in survival was found, suggesting that presence of bone metastasis is a negative prognostic factor independent of treatment type. Of note, liver metastasis was also identified as a negative prognostic factor in that study[21]. In another study of patients with aUC treated with 2nd+ line ICI, Raggi et al. showed that bone metastasis was associated with significantly shorter OS and PFS[22].

Similarly, another study by de Liano Lista et al. showed that concurrent presence of bone and liver metastases, along with multi-metastatic disease (defined as ≥ 3 sites) was associated with shorter OS in

patients with aUC treated with ICIs in the 1st line setting. The association persisted in the 2nd line, while the presence of liver-only and bone-only metastasis was significantly associated with shorter OS, while LN-confined metastasis was associated with longer OS[23]. Moreover, Ma et al. assessed 160 patients with urothelial or renal cell carcinoma treated with ICI and showed that the presence of liver metastasis was associated with significantly worse ORR, PFS and OS. Of interest, comparing liver to LN-confined metastasis, CNS or bone metastasis showed that patients with liver metastasis had worse outcomes[24].

In our study, we also investigated the putative association between lung metastasis and outcomes with ICI; interestingly, presence of lung, without liver or bone metastases, was significantly associated with shorter PFS only in the 2nd+ line setting. Lung metastasis was associated with worse outcomes when co-present with liver and/or bone metastases. Results may suggest that lung-confined metastasis may not have the same magnitude as a negative prognostic factor in patients with aUC treated with ICI, but more studies are needed to evaluate this hypothesis.

Subgroup analyses from clinical trials evaluating ICI in aUC are aligned with our findings. In the IMvigor 210 trial, ORR with atezolizumab was 23% (95% CI:16-31) for the entire cohort 1 (consisting of cisplatin-ineligible patients without prior treatment for aUC), but numerically higher in patients with LN-confined metastasis (n=31; 32% [95% CI:17-51]) and lower for those with liver metastases (n=25, 8% [95%CI: 1-26]). Median OS in patients with liver metastasis was also shorter (5.5 vs 15.9 months in all patients)[12]. In the Keynote 052 trial, patients with LN-confined metastasis (n=51) had higher ORR on pembrolizumab than those with visceral metastases (n=315): 47% (95% CI:31-62) vs 23% (95% CI:18-29).[13] In the Keynote 045 trial, while the HR for OS was <1 (favoring pembrolizumab vs salvage chemotherapy) for both patients with and without liver metastasis, HR for those with liver metastasis was higher (0.85 vs 0.67), which might suggest lower degree of benefit in this population, though the trial was not powered for these subgroup analyses.[14]

Although the above subset analyses were purely exploratory and should not be over-interpreted due to the small sample size and number of events, they raise the hypothesis of worse outcomes in patients with non-LN metastases treated with ICI. These results in the context of our findings suggest that the spread of tumors beyond LNs may possibly herald a shift in cancer biology and/or suggest that the LN-confined metastasis could possibly be more sensitive to ICI due to a strong presence of immune cells in LNs. However, it is important to note that visceral (liver, lung or bone) metastasis appear as a negative prognostic factor since patients seem to have worse outcomes regardless of therapy, e.g. chemotherapy or ICI. Moreover, there are not adequate datasets to clarify the putative predictive role of metastatic site with regards to response and survival to individual therapies. The latter 'predictive' question requires further analysis in randomized trials and cannot be answered in single arm trials and retrospective studies.

It is unclear whether the above findings could be attributed to underlying cancer biology/organotropism, such as increased clonal evolution, tumor heterogeneity, cancer burden, tumor mutation burden, quality of neoantigens, PD-L1 or other factors in the tumor microenvironment, and/or host-related factors, such as immunosuppression, T cell clonality and diversity, and other components of immune response[25,26]. Emerging evidence also suggests that differences exist on immune-related factors, such as tumor-infiltrating lymphocytes and PDL1 expression on immune cells between primary tumors and metastatic sites[27]. Such immune heterogeneity across metastatic sites could pose significant treatment challenges. Disease heterogeneity seems to extend to a molecular level as well; recent work showed that MTAP-deficient tumors seem significantly more likely to present with visceral metastasis, such as liver or lung, and are associated with worse prognosis with ICI[28]. Another study on heterogeneity following chemotherapy showed that chemotherapy-treated aUC had increased mutational variation between different clones, while clonal expansion of primary tumors happened early in the disease course[29]. Similarly, a recently developed model/score of clinical (NLR ≥5, visceral metastasis) and molecular factors (Single-Nucleotide Variant, SNV <10) correlated with no benefit with ICI in patients with aUC[11].

Such findings require further validation in larger cohorts; they, however, may suggest a distinct molecular background associated with early visceral metastasis and poor outcomes with ICI, and could be useful in deciding optimal sequencing of ICI therapy in the future.

Notably, in addition to ICI, other agents, such as enfortumab-vedotin, erdafitinib and sacituzumab-govitecan have been approved by the FDA for treatment-refractory aUC. For patients with FGFR2 or FGFR3 activating mutation or fusion, the BLC 2001 single arm trial with erdafitinib showed an ORR 40% in patients with progression on at least one prior course of chemotherapy. In this study, outcomes were similar among patients with liver (ORR 35% [95% CI: 14-56]) and bone metastasis (ORR 48% (95% CI: 26-69) compared to the overall population [4]. Similarly, in a subgroup analysis of the EV201 trial with 2nd line enfortumab-vedotin in cisplatin-ineligible patients previously treated with ICI, outcomes were similar between patients with or without liver metastasis; ORR in patients with liver lesions was 48% (95% CI: 26-70) and 53% (95% CI: 40-65) for those without[30]. In the EV301 trial of EV for patients previously treated with platinum-based chemotherapy and ICI, HR for OS in patients with liver metastasis (HR: 0.66, 95% CI: 0.46-0.96) was similar to the HR for those without liver metastasis (HR: 0.73 95% CI:0.55-0.98). Similarly, in a real-world study of patients with aUC treated with enfortumab-vedotin (UNITE), patients with liver metastases demonstrated a higher ORR (64% vs 47%, p<0.05), but shorter OS (8.3 months, vs 15.7 months, p=0.05) compared to those without liver metastasis; the presence of bone metastases was not associated with a significant difference in ORR or OS in the UNITE study[21].

In the context of the above separate datasets, a potential application of our findings could be to help inform treatment decisions among patients with aUC based on metastatic sites[31]. For patients with bone and/or liver metastases, our data suggests poor response and survival with ICI. Recently, enfortumab-vedotin received FDA approval after one or more prior lines of therapy in cisplatin-ineligible patients. Moreover, the THOR (NCT03390504) phase III trial is comparing erdafitinib to pembrolizumab in patients with platinum-refractory aUC harboring a susceptible FGFR alteration (results are pending). So far, there has been no head-to-head comparison between either antibody drug conjugate or erdafitinib vs ICI in the platinum-refractory setting. Based on all the above considerations, therapeutic agents with higher ORR in patients with liver and/or bone metastasis could be possibly considered prior to use of ICI in this setting.

Treatment with ICI may depend more on the immune system robustness compared to cytotoxic regimens; patients with advanced cancer may have less robust immune system, which might dampen response to ICI[32]. Recent trials with neoadjuvant ICI monotherapy (e.g. ABACUS, PURE-01) have showed promising results with high pathologic complete response rates[33]. Moreover, there have been practice-changing data in the adjuvant and switch maintenance settings based on the Checkmate 274 and Javelin Bladder 100 phase III trials[34,35]. These findings, along with data on the limited efficacy of single-agent ICI in patients with visceral metastases and higher tumor burden might suggest that ICI used in earlier disease settings could possibly have greater benefit compared to the salvage setting. Several clinical trials have also investigated the role of ICI alone or combined with other systemic therapies and/or radiation in earlier disease settings, with promising results[36–39]. However, this hypothesis is not confirmed, while the role of ICI is evolving in UC.

Our work has limitations inherent to its retrospective design, such as lack of randomization, possibility for selection bias, and residual confounding that cannot be measured or fully accounted for. Clinical practices, surveillance schedules and follow-up times may not be perfectly consistent across 26 different institutions. Additionally, no centralized review of pathology and imaging was feasible, but all participating sites were academic institutions with expert genitourinary medical oncologists, radiologists and pathologists. Although we tried to capture information on the extent of metastatic burden, detailed data on the number of distinct lesions per organ system, as well as their size, were not available. Sample size in many of the metastatic subgroups was also limited by low power (~10-30 patients). Response and

progression to treatment were determined by systematic comprehensive chart review based upon clinical and radiological notes, while prespecified interval assessments using strictly the RECIST 1.1 criteria were not mandated. We did not capture data on switch maintenance ICI or on molecular biomarkers. However, our study has several strengths, including the utilization of real-world data outside clinical trials, a relatively large sample size, and participation of multiple institutions across two continents with a diverse patient population.

In conclusion, we showed that in patients receiving ICI for aUC outside clinical trials, the presence of bone and/or liver metastasis was significantly associated with lower ORR, as well as shorter PFS and OS. Despite inherent limitations, these results are complementary to findings from relatively limited trial subset analyses and could possibly help inform therapeutic discussions, prognostic estimates, as well as the design, stratification, and interpretation of clinical trials with ICI in aUC. Future research is needed to further identify and validate not only prognostic, but also predictive, biomarkers that can help inform optimal use of ICI in patients with aUC.

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Tables & Figures:

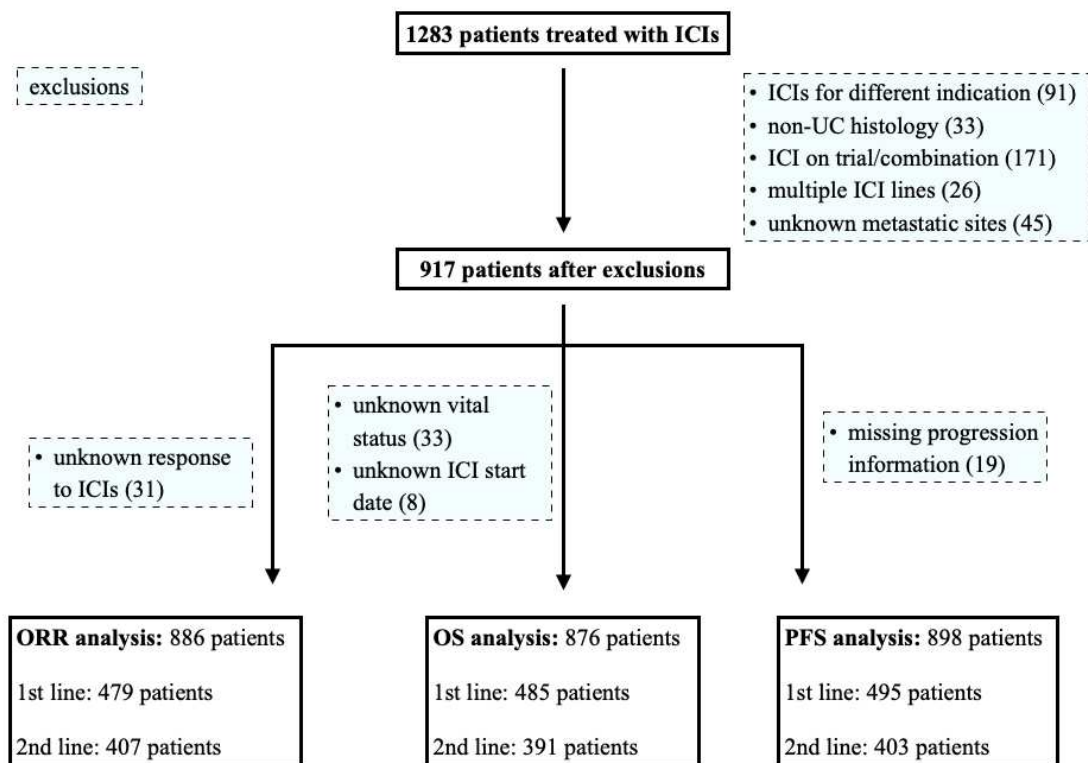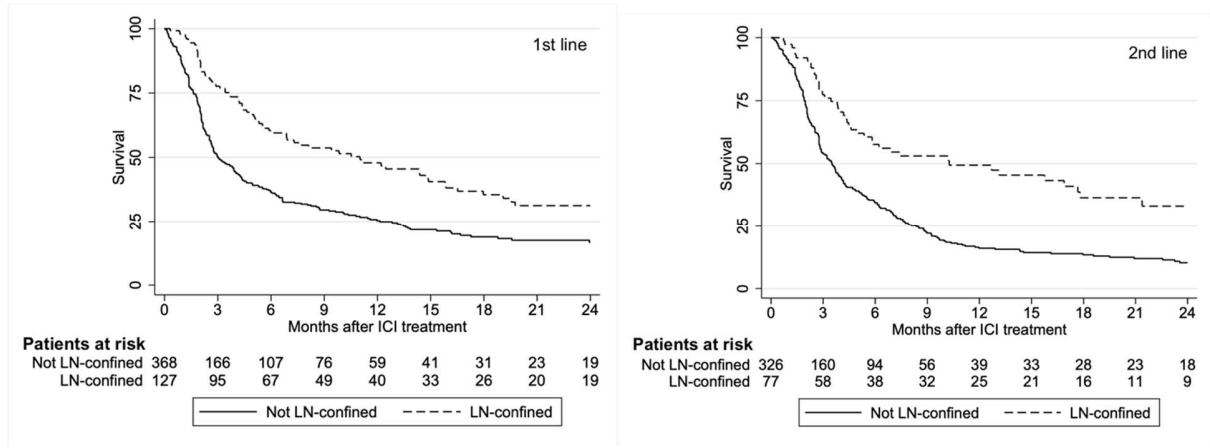**Figure 1: Consort diagram**

Figure 1: Consort flow diagram of patient selection and exclusion rationale

Figure 2: Progression-Free Survival

2a. Lymph Node (LN) model (model A)
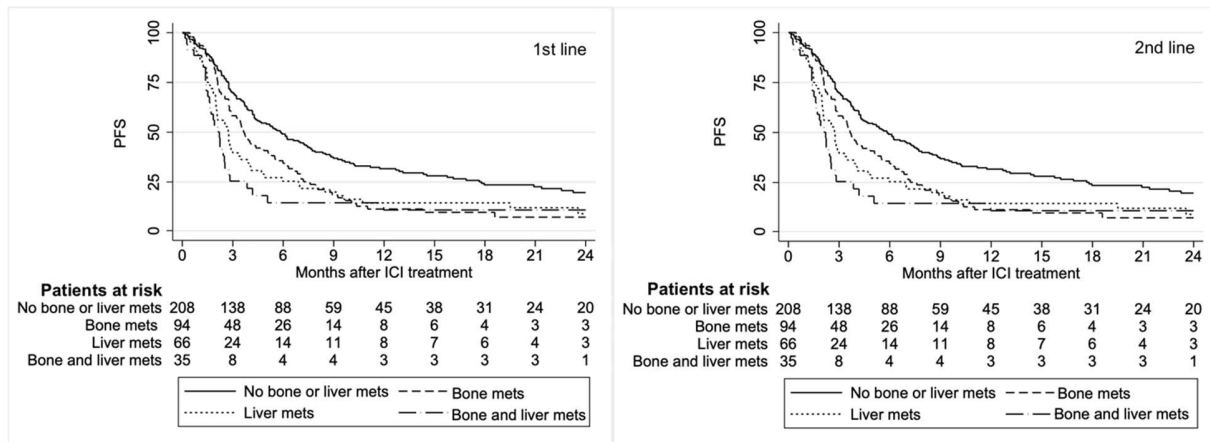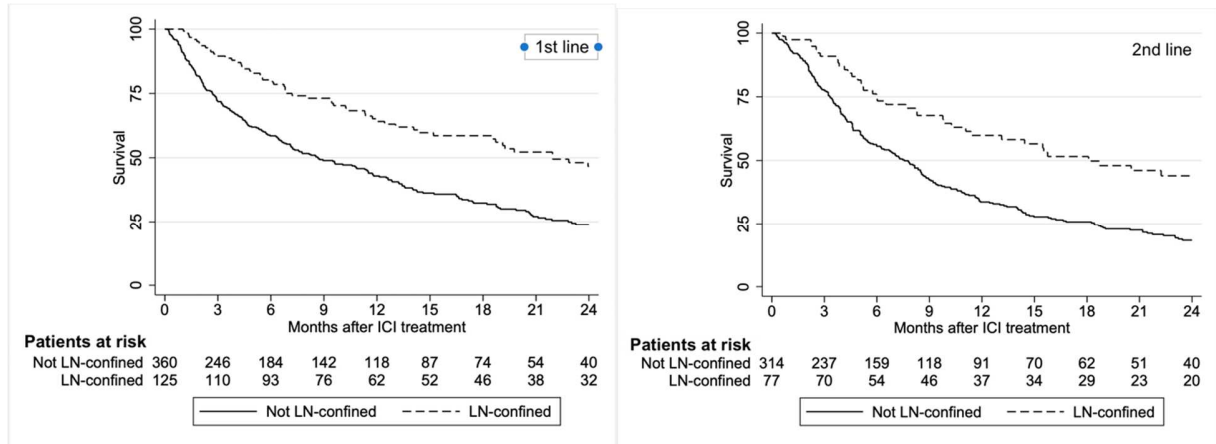

2b. Liver & Bone model (model B)

Figure 3: Overall Survival

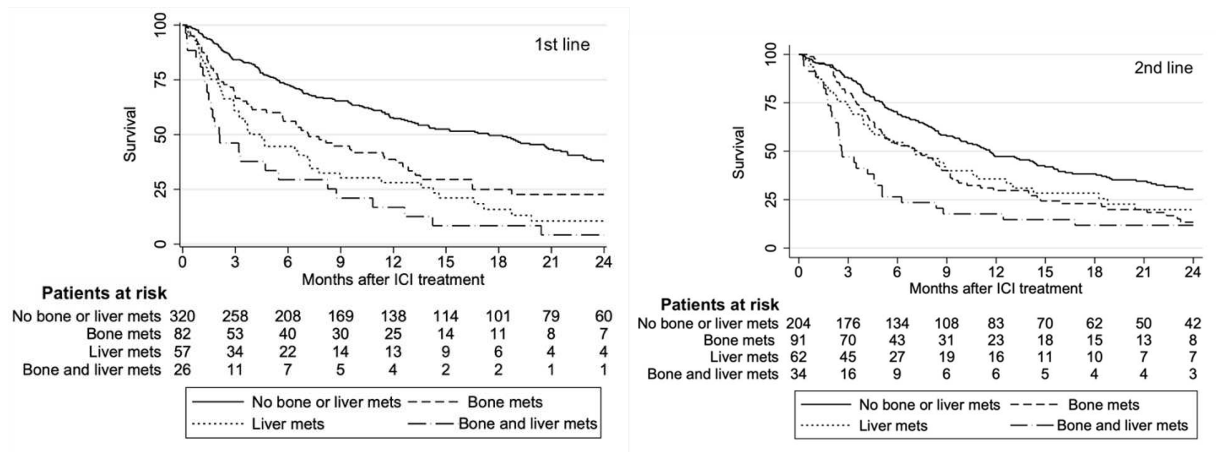### 3a. Lymph node (LN) model (model A)

3b. Liver & Bone model (model B)

Table 1: Patient population demographics

		First Line ICI N = 504	Subsequent Line ICI N = 413
Age at ICI initiation		71 (11)	68 (10)
Sex	Male	360 (71%)	313 (76%)
	Female	144 (29%)	100 (24%)
Smoking History	No	162 (32%)	136 (33%)
	Yes	339 (68%)	273 (67%)
White race	no	109 (22%)	89 (22%)
	yes	395 (78%)	324 (79%)
History of cystectomy or (nephro)ureterectomy	No	210 (44%)	192 (50%)
	Yes	268 (56%)	191 (50%)
Histology	Pure Urothelial	344 (69%)	302 (74%)
	Mixed Urothelial	158 (32%)	108 (26%)
Hemoglobin <10mg/dl at ICI initiation	No	371 (75%)	291 (72%)
	Yes	124 (25%)	113 (28%)
Prior platinum chemotherapy	No	303 (60%)	29 (7%)
	Yes	201 (40%)	384 (93%)
ECOG PS at ICI initiation	0	121 (26%)	93 (25%)
	1	211 (46%)	210 (57%)
	2	112 (24%)	53 (14%)
	3	14 (3%)	12 (3%)
	4	2 (<1%%)	0 (0%)
Bellmunt score at ICI initiation	0	90 (20%)	63 (17%)
	1	228 (50%)	166 (46%)
	2	117 (26%)	110 (30%)
	3	17 (4%)	25 (7%)
Site of Primary Tumor	Bladder	410 (83%)	316 (79%)
	Upper GU	25 (5%)	25 (6%)
	Urethra	5 (1%)	4 (1%)
	Renal Pelvis	39 (8%)	33 (8%)
	Ureter	14 (3%)	24 (6%)
Lymph node metastasis at ICI initiation	no	143 (28%)	88 (21%)
	yes	361 (72%)	325 (79%)

Lung metastasis at ICI initiation	no	342 (68%)	251 (61%)
	yes	162 (32%)	162 (39%)
Liver metastasis at ICI initiation	no	420 (83%)	312 (76%)
	yes	84 (17%)	101 (25%)
Bone metastasis at ICI initiation	no	394 (78%)	282 (68%)
	yes	110 (22%)	131 (32%)

Table 2: Lymph Node metastatic disease model:

	Metastatic sites	ORR	OR	PFS	HR	OS	HR
1ST line ICI	Non LN - confined disease	22 (18-26)	reference	3 (3-4)	reference	9 (7-12)	reference
	LN - confined disease	45 (37-54)	**2.97 (1.92-4.59)***	11 (7-16)	**0.53 (0.41-0.69)***	22 (14-NR)	**0.49 (0.37-0.65)***
2nd line ICI or greater							
	Non LN - confined disease	19 (15-23)	reference	4 (3-4)	reference	8 (6-9)	reference
	LN - confined disease	38 (28-49)	**2.79 (1.50-5.16) *#**	13 (6-21)	**0.47 (0.34-0.65)***	18 (11-NR)	**0.54 (0.38-0.77)*β**

*: p<0.05
#: adjusted for cystectomy and albumin at ICI initiation
β: adjusted for albumin

Table 3: Liver and bone metastatic disease model:

	Metastatic sites	ORR	aOR	PFS	aHR	OS	aHR
1st line ICI							
	No liver-bone	33 (28-39)	Reference	9 (6-13)	Reference	17 (13-21)	Reference
	Bone - only	21 (14-32)	**0.48 (0.25-0.91)*#**	3 (2-6)	**1.65 (1.19-2.30)*β**	7 (4-11)	**1.60 (1.13-2.25)*γ**
	Liver - only	15 (7-28)	**0.31 (0.13-0.75)*#**	2 (2-3)	**2.54 (1.77-3.66)*β**	5 (3-7)	**2.35 (1.66-3.34)*γ**
	Liver & bone	4 (1-22)	**0.08 (0.01-0.06)*#**	2 (1-3)	**3.23 (1.96-5.32)*β**	2 (1-5)	**3.66 (2.31-5.79)*γ**
2nd line ICI or greater							
	No liver-bone	28 (23-35)	Reference	6 (5-8)	Reference	12 (9-15)	Reference
	Bone - only	12 (7-20)	**0.29 (0.13-0.67)*δ**	4 (3-5)	**1.71 (1.28-2.28)*ε**	7 (5-9)	**1.76 (1.32-2.35)*ε**
	Liver - only	25 (16-37)	0.70 (0.34-1.45) δ	3 (2-6)	**1.62 (1.09-2.41)*ε**	7 (4-11)	**1.56 (1.04-2.34)*ε**
	Liver & Bone	11 (4-27)	0.45 (0.15-1.39) δ	2 (2-3)	**2.32 (1.34-4.03)*ε**	3 (2-5)	**2.53 (1.47-4.38)*ε**

*: p<0.05
#: adjusted for cystectomy, site of primary (upper vs lower) and albumin at ICI initiation
β: adjusted for albumin at ICI initiation
γ: adjusted for ECOG PS at ICI initiation
δ: adjusted for cystectomy, hemoglobin, albumin and NLR at ICI initiation

Table 4: Liver, bone and lung metastasic disease model:

*: p<0.05

	Metastatic sites	ORR	aOR	PFS	aHR	OS	aHR
1st line ICI							
	No liver-bone-lung	37 (31-43)	Reference	7 (5-11)	Reference	17 (12-22)	Reference
	Bone - only	20 (12-33)	0.48 (0.23-1.01)#	3 (2-5)	1.42 (0.93-2.16)$^{\alpha}$	7 (3-13)	1.55 (0.96-2.51)$^{\beta}$
	Liver - only	13 (5-29)	**0.21 (0.06-0.72)*#**	2 (1-3)	**2.43 (1.55-3.83)*$^{\alpha}$**	5 (3-14)	**2.46 (1.54-3.94)*$^{\beta}$**
	Lung-only	26 (18-35)	0.69 (0.40-1.21)#	4 (3-6)	1.32 (0.96-1.83)$^{\alpha}$	18 (11-23)	1.25 (0.87-1.79)$^{\beta}$
	Liver & bone	6 (1-32)	0.14 (0.02-1.04)#	1 (1-4)	**2.09 (1.08-4.05)*$^{\alpha}$**	2 (1-5)	**2.81 (1.46-5.37)*$^{\beta}$**
	Lung & bone	24 (10-46)	0.31 (0.09-1.11)#	4 (1-12)	1.39 (0.71-2.74)$^{\alpha}$	11 (3-NR)	1.59 (0.80-3.14)$^{\beta}$
	Lung & liver	19 (6-45)	0.33 (0.07-1.65)#	2 (2-3)	**2.52 (1.40-4.55)*$^{\alpha}$**	4 (2-7)	**3.74 (2.11-6.62)*$^{\beta}$**
	Lung, liver & bone	0	-	2 (0.2-2)	**3.92 (1.77-8.70)*$^{\alpha}$**	2 (0.2-13)	**4.41 (2.19-8.88)*$^{\beta}$**
2nd line ICI or greater							
	No liver-bone	32 (25-41)	Reference	7 (5-10)	Reference	14 (11-19)	Reference
	Bone - only	14 (7-25)	**0.25 (0.09-0.72)*$^{\gamma}$**	4 (3-7)	**1.75 (1.17-2.63)*$^{\delta}$**	8 (5-10)	**1.87 (1.24-2.82)*$^{\varepsilon}$**
	Liver - only	24 (13-39)	0.49 (0.20-1.23)$^{\gamma}$	3 (2-4)	**2.20 (1.34-3.59)*$^{\delta}$**	9 (4-14)	1.61 (0.93-2.80)$^{\varepsilon}$
	Lung-only	22 (15-32)	0.67 (0.33-1.36)$^{\gamma}$	4 (3-6)	**1.58 (1.10-2.28)*$^{\delta}$**	8 (6-12)	1.33 (0.90-1.97)$^{\varepsilon}$
	Liver & Bone	9 (2-30)	0.28 (0.05-1.52)$^{\gamma}$	2 (1-3)	**2.91 (1.30-6.52)*$^{\delta}$**	2 (2-5)	**2.80 (1.21-6.44)*$^{\varepsilon}$**
	Lung & bone	8 (3-23)	**0.25 (0.07-0.87)*$^{\gamma}$**	3 (2-5)	**2.49 (1.72-3.61)*$^{\delta}$**	6 (4-9)	**1.96 (1.31-2.95)*$^{\varepsilon}$**
	Lung & liver	26 (12-47)	1.13 (0.34-3.83)$^{\gamma}$	3 (2-3)	**2.34 (1.27-4.33)*$^{\delta}$**	4 (3-9)	**2.14 (1.14-4.02)*$^{\varepsilon}$**
	Lung, liver & bone	15 (4-45)	0.60 (0.13-2.80)$^{\gamma}$	2 (1-3)	**2.64 (1.24-5.60)*$^{\delta}$**	3 (1-9)	**2.78 (1.26-6.15)*$^{\varepsilon}$**

#: adjusted for cystectomy, site of primary (upper vs lower), albumin at ICI initiation and NLR at ICI initiation

$^{\alpha}$: adjusted for cystectomy, site of primary (upper vs lower), and albumin at ICI initiation

$^{\beta}$: adjusted for cystectomy, site of primary (upper vs lower), ECOG at ICI initiation, albumin, hemoglobin and NLR at ICI initiation

$^{\gamma}$: adjusted for cystectomy, site of primary (upper vs lower), albumin, hemoglobin and NLR at ICI initiation

$^{\delta}$: adjusted for cystectomy, albumin, hemoglobin and NLR at ICI initiation

$^{\varepsilon}$: adjusted for cystectomy, ECOG at ICI initiation, albumin, hemoglobin and NLR at ICI initiation