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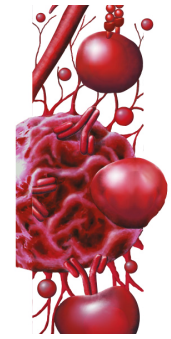

The immunotherapy revolution in genitourinary malignancies

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Immune checkpoint inhibitor (ICI) therapy and therapeutic cancer vaccines have continued to demonstrate survival benefit and durable clinical response in patients with renal cell cancer, prostate cancer and bladder cancer, with limited responses in testicular cancer. The role of immunotherapy in combination with chemotherapy or other targeted therapies in the neo-adjuvant, adjuvant and metastatic setting is actively being explored. We describe the current immunotherapy-related treatment modalities approved for genitourinary cancers, focusing on immune checkpoint inhibitors, vaccines and other modalities, and highlight ongoing studies involving immunotherapy in these cancer types.

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In the past decade, immunotherapy has dramatically impacted the treatment of patients with cancer. Active forms of immunotherapy include tumor antigen-directed vaccines and immune checkpoint inhibitors (ICIs). Of the ICIs shown to have clinical efficacy, CTLA-4, PD-1 and PD-1 ligand (PD-L1) directed therapy has proven to be most beneficial in patients including those with malignant melanoma [1], non–small-cell lung cancer, renal cell cancer, with an objective response rate (ORR) of up to 20% and durability of responses lasting over 1 year [2–4].

CTLA-4 and PD-1 and their corresponding ligands B7-1/B7-2 and PD-L1 play an important role in dampening T-cell activity and evading the immune checkpoint pathway, thereby minimizing T-cell response to cancer-associated antigen. Cancer cells can overexpress these proteins and evade a tumor-directed immune response by negatively regulating the immune system. Monoclonal antibodies (mAbs) against CTLA-4 (ipilimumab), PD-1 (nivolumab and pembrolizumab), or PD-L1 (avelumab, atezolizumab, and durvalumab) can stimulate the immune system by reactivating T cells and result in clinical responses described above. PD-L1 expression has been shown to correlate with response rates in some tumor types but not others denying it a clear role as a predictive biomarker across all malignancies. Interestingly, mismatch repair status and microsatellite instability predict clinical benefit of immune checkpoint blockade [5,6] and as a result, pembrolizumab, an anti-PD-1 agent, has been approved as second-line therapy for solid tumors with mismatch repair deficiency in a tissue agnostic fashion. With the evolving use of immunotherapy, the care for patients with genitourinary malignancies has been significantly impacted and it is likely that this evolution will increase our understanding of these therapies.

Renal cell carcinoma

Early & localized renal cell carcinoma

Renal cell carcinoma (RCC) is managed based on disease extent and histology. Localized RCC includes disease limited to the kidney and advanced disease with extension into the major veins, perinephric tissues, which may be managed surgically. However, more advanced RCC or metastatic disease that is not resectable may be more amenable to systemic therapy.

There is no clear role for immunotherapy in localized disease or for adjuvant immunotherapy after definitive local therapy of advanced disease, though studies are currently ongoing. Multiple studies have shown no clear benefit using older generation immunotherapy drugs such as IFN-α, IL-2 and autologous tumor vaccines as ad-

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juvant therapy after resection of localized or locally advanced RCC. However, there are multiple ongoing trials evaluating the efficacy of various combinations of immunotherapy in localized RCC and in various settings including adjuvant nivolumab and ipilimumab vs placebo (CheckMate-914; NCT NCT03138512), Nivolumab alone (NCT03055013), perioperative nivolumab neoadjuvant followed by nephrectomy and further adjuvant nivolumab for 6 months (PROSPER; NCT03055013), pembrolizumab (MK-3475-564/KEYNOTE-564; NCT03142334), atezolizumab (IMmotion010; NCT03024996) and neoadjuvant avelumab with axitinib followed by surgical resection (NEOAVAX trial, NCT03341845), an open-label, single-arm, Phase II trial with the primary end point of response via RECIST 1.1 (Response Evaluation Criteria In Solid Tumors) following neoadjuvant therapy [7].

Prior to the modern era of immunotherapy heralded by ICIs, cytokines had demonstrated efficacy in RCC. Clinical trials demonstrated IFN-α monotherapy at escalating doses compared with medroxyprogesterone acetate resulted in OS of 43% (vs 31% medroxyprogesterone acetate) at 1-year interval and a 28% risk reduction of death with median survival of 8.5 months (vs 6 months with medroxyprogesterone acetate) [8], though use of these agents have largely fallen out of favor. Patients with prior nephrectomy have been shown to derive a survival benefit with IFN-α, with post-nephrectomy IFN-α treatment prolonging median survival when compared with patients who received IFN-α without nephrectomy (~11 months vs 8.1 months) [9,10]. Several large trials have evaluated the use of monotherapy with IFN-α in patients with metastatic RCC. A meta-analysis of these trials demonstrated median OS of 13 months and median time to progression of 4.7 months, with higher response times correlating directly with risk stratification. The favorable-risk group had a 20-month median survival time compared with 14 months for intermediate-risk group and 5 months for poor-risk group [11].

Treatment with high-dose IL-2 can boost immune response in certain subset of patients with metastatic RCC. Treatment is associated with severe toxicity such as hematologic toxicities, capillary leak syndrome, CNS toxicity and a relatively low ORR of 20% (9% CR and 11% PR) and the response was durable [12,13]. However, there remains a small subgroup of patients who truly benefit from high-dose IL-2 as seen in the PROCLAIM registry dataset, even beyond treatment of PD-1/PD-L1 inhibitors [14].

In a study comparing the use of IL-2, IFN-α, or combination in metastatic RCC, the overall responses reflected prior studies; best response rate was 19% with combination treatment with both IL-1 and IFN-α, 20% event-free survival at 1 year, but no change in OS. Severe toxicities were more common with IL-2 than with IFN-α [15]. Efforts to capitalize on the benefits of IL-2 have come in the form of refining IL-2 targeting such as by using bempegaldesleukin (NKTR-214), a CD122-preferential IL-2 pathway agonist that stimulates proliferation and activation of tumor antigen-specific $CD8^+$ T cells and natural killer cells within the tumor microenvironment, which can thereafter increase PD-1/PD-L1 expression and is being studied now in varying combinations in mRCC (NCT03729245). However, given better survival benefits and lower toxicity/side effects, the use of IFN-α and IL-2 has largely been replaced by ICI therapy in the first-line setting.

Advanced & metastatic clear cell RCC

Advanced and unresectable RCC can be treated with immunotherapy based on risk stratification and classification by clear cell versus non-clear cell histology, although most trials included clear cell histology which makes up the most common subtype of all renal cell carcinomas. Immunotherapy approaches that have been proven to be efficacious in this setting (see Table 1) includes the use of ICI and IL-2.

For patients who present with untreated locally advanced or metastatic clear cell RCC, surgery is not expected to render a cure. Patients with clear cell RCC are initially risk-stratified using the IMDC (International Metastatic RCC Database Consortium) risk score, which is calculated using their Karnofsky Performance Score (KPS), time from diagnosis to initiation of systemic therapy, hemoglobin, calcium, neutrophil count and platelet count. The role of cytoreductive nephrectomy continues to evolve given results of a recent Phase III clinical trial (CARMENA) which revealed that patients with intermediate- and poor-risk disease may not benefit from upfront surgery and benefit instead from systemic therapy with sunitinib [16]. For patients with intermediate-risk disease (defined as 1–2 risk factors) or poor-risk disease (IMDC score >3), first-line therapy consists of the combination of ipilimumab (1 mg/kg) combined with nivolumab (3 mg/kg) given intravenously every 3 weeks for four cycles then continued with maintenance nivolumab every 2 or 4 weeks until disease progression per clinician's choice. This is based on the pivotal randomized Phase III clinical trial Checkmate-214, which demonstrated increased OS in intermediate and poor risk groups with advanced/metastatic RCC compared with sunitinib, with an 18-month OS of 75% and ORR of 42% (vs. OS of 60% and ORR of 27% in sunitinib group) [17]. The median OS for the ipilimumab with nivolumab group was not reached and was 26 months with the sunitinib group. The median PFS was

Table 1. Phase III trials using US FDA-approved immune checkpoint inhibitors in metastatic renal cell carcinoma.

Trial name	Drug mechanism of action	Drug dosages and experimental arms (number of patients)	Primary end points	Secondary end points	Results of primary end point
CheckMate-214	Ipilimumab CTLA-4 inh. + Nivolumab PD-1 inh. vs sunitinib VEGF TKI	Nivolumab 3 mg/kg IV + Ipilimumab 1 mg/kg IV q 3 weeks × 4 (Induction) then maintenance Nivolumab vs sunitinib 50 mg daily (4 weeks on/2 weeks off) (n = 1096)	ORR, PFS and OS in intermediate-risk and poor-risk	ORR, PFS, OS in intent-to-treat population	Nivo + Ipi OS = NR vs sunitinib = 26.0 months
JAVELIN Renal 101	Avelumab PD-L1 inh. + axitinib vs sunitinb VEGF TKI	Avelumab 10 mg/kg IV q 2 weeks + Axitinib 5 mg BID (6-week cycle) vs sunitinib 50 mg daily (4 weeks on/2 weeks off) (n = 886)	PFS, OS in PD-L1+ (63.2% of tumors)	PFS in ITT; ORR, treatment-related AEs	mPFS Avelumab + axitinib = 13.8 mos vs sunitinb = 7.2 mos
KEYNOTE-426	Pembrolizumab PD-1 inh. + axitinib vs sunitinib VEGF TKI	Pembrolizumab 200 mg IV q 3 weeks + axitinib 5 mg BID (6-week cycle) vs sunitinib 50 mg daily (4 weeks on/2 weeks off) (n = 840)	PFS, OS in ITT	ORR, DCR, DOR, PFS, OS, AEs	12-mo OS Pembrolizumab + axitinib = 89.9% vs. 78.3% sunitinib group; mPFS Pembrolizumab + axitinb = 15.1 mos vs sunitinb = 11.1 mos
CheckMate-025	Nivolumab PD-1 inh vs Everolimus mTOR inhibitor	Nivolumab 3 mg/kg IV q 2 weeks vs sunitinib 50 mg daily (4 weeks on/2 weeks off) (n = 821)	PFS	ORR, safety	mOS Nivolumab = 25.0 mos vs everolimus = 19.6 months

BID: Twice a day; DOR: Duration of response; Hazard ration: Hazard ratio; HRQOL: Health-related quality of life; ICI: Immune checkpoint inhibitor; inh: Inhibitor; ITT: Intention-to-treat population; mRCC: Metastatic renal cell carcinoma; n: Number; ORR: Objective response rate; OS: Overall survival; PD-L1: PD-1 ligand; PFS: Progression-free survival; q: Every; TKI: Tyrosine kinase inhibitor.

11.6 months for the ipilimumab and noivolumab group vs. 8.4 months for the sunitinib group but not statistically significant. Both ORR and PFS were higher and seemed more beneficial in patients with PD-L1 expression ≥1%. However, since responses were also seen in those without PD-L1 expression, there was no limitation in terms of use of ipilimumab and nivolumab based on PD-L1 biomarker expression. However, further post-hoc analysis showed that the response rate was lower (29 vs 52%) and PFS was shorter (median 15.3 vs 25.1 months) for patients with favorable disease compared with sunitinib [18,19], hence limiting the US FDA label indication to patients with intermediate- or poor-risk disease [20]. Toxicity profile with combination immunotherapies yield expected autoimmune side effects which led to treatment discontinuation in 22% of patients in the combination nivolumab and ipilimumab arm compared with 12% in the sunitinib arm, with requirement of use of any steroids at 60%. The effects of monotherapy with ipilimumab or nivolumab and its efficacy in treatment-naive RCC are not clear. However, given the knowledge of toxicities associated with ipilimubab from previous studies, there are ongoing trials evaluating the use of initial single-agent nivolumab in advanced and metastatic RCC with addition of ipilimumab only in patients who do not have response to or progress on nivolumab monotherapy (NCT02210117; NCT02917772; NCT03873402; NCT03177239; NCT03203473).

Brain metastases occur in about 10% of RCC patients and these patients are initially treated with surgery and/or radiation prior to systemic therapy due to risk of hemorrhage with untreated tumors [21]. Data on the efficacy of ICIs in this subgroup of patients is lacking, as these patients are often excluded from clinical trials. However, recent data from the open-label CheckMate-920 trial, which was a Phase IIIb/IV trial with several cohorts that included a brain metastases cohort, suggests efficacy of ipilimumab with nivolumab in this subgroup as well [22]. Patients treated for four cycles with ipilimumab and nivolumab followed by maintenance nivolumab in patients with brain metastases, with a PFS of 9 months and ORR of 29%. It appears that nivolumab monotherapy provides marginal benefit in radiation-naive patients with brain metastases. In one open-label study, Phase II study, nivolumab monotherapy resulted in PFS of 2.7 months and an ORR with decrease in intracranial metastatic sites of 12% in radiation-naive RCC patients with brain metastases who experienced disease progression on antiangiogenic therapy compared with patients previously treated with radiation, who had no objective responses [23]. Despite the slight improvement in response and PFS, 72% of the radiation-naive patients eventually required subsequent locoregional radiation for progressive disease.

Combining ICI with targeted therapies in RCC has also been explored and has become an additional standard-of-care as first-line therapy in locally-advanced or metastatic RCC in the form of JAVELIN Renal 101 and KEYNOTE-426 trials. The combination using PD-L1 inhibitor avelumab with VEGF inhibitor axitinib was evaluated in the Phase III JAVELIN Renal 101 trial, which did show the combination of avelumab with axitinib

improved PFS relative to sunitinib (13.8 vs 7.2 months) but did not demonstrate an OS benefit as the data was immature at the time of publication [24]. Regardless, this garnered FDA approval also in 2019.

KEYNOTE-426 evaluated the combination of pembrolizumab and a VEGF inhibitor axitinib as a first-line therapy for patients with untreated advanced or metastatic RCC, compared with the known standard-of-care at the time of sunitinib, demonstrating improved OS (90 vs 78% at 12 months), longer PFS (15.1 vs 11.1 months), better ORR (59 vs 36%), lower side effects and adverse events when compared with sunitinib [25]. These benefits were seen regardless of IMDC risk classification. This trial has led to the FDA approval of the combination of pembrolizumab and axitinib in 2019 [26]. However, there are patients who are not able to tolerate or eventually have to stop or dose-reduce the axitinib. For these patients, data to support use of single agent pembrolizumab exists and was studied in the KEYNOTE-427 trial with pembrolizumab monotherapy [27]. The trial demonstrated an ORR of 34% at a median follow-up of 12 months with higher response rate (38%) in intermediate- and poor-risk disease with PD-L1 expression >1%.

The combination of another PD-L1 inhibitor atezolizumab and bevacizumab has also been studied in the IMmotion 151 trial. Atezolizumab and bevacizumab has been proven to be superior when compared with sunitinib and a Phase II trial demonstrated longer PFS (11.2 vs 8.4 months), and better ORR (37 vs 33%), and higher complete response (CR) rate (5 vs 2%) [28]. The median PFS was longer (11.2 vs 7.7 months) and the ORR was higher (43 vs 35%) in patients who had a PD-L1 ≥1% but the OS was not significantly different in either group [28]. This has not yet garnered FDA approval for use as first-line treatment.

Despite the benefits demonstrated by each of these combinations, questions remain as to whether or not sunitinib, considered the de facto standard-of-care at the time, was the optimal control arm for many of the studies, since the standard-of-care has now clearly rapidly evolved. Furthermore, questions remain about how these different, potentially front-line regimens compare to each other and if immunotherapy and targeted therapy combinations are better than those agents given sequentially.

Relapsed or progressive metastatic RCC: role of second-line therapy

Progression on any initial ICI therapy requires further treatment with anti-VEGF or TKI such as axitinib, cabozantinib, sunitinib or pazopanib. If patient has received axitinib with avelumab or ipilimumab with nivolumab, non-ICI therapy such as lenvatinib or everolimus remain viable options, though optimal sequencing remains unknown.

For patients who have progressive disease while on anti-VEGF agent such as axitinib, cabozantinib, sunitinib, or pazopanib, nivolumab monotherapy remains a second-line treatment option, based on the randomized Phase III CheckMate-025 trial [29]. When compared with everolimus alone, treatment with nivolumab resulted in significantly longer OS (25 vs 20 months; HR: 0.73) and higher ORR (25 vs 5%) with less grade 3 or higher toxicity. Interestingly none of the benefits were associated with higher PD-L1 expression.

For patients who progress on or after being treated with the combination of pembrolizumab and axitinib, second-line treatment with ipilimumab with nivolumab has been used in clinical practice as an off-label treatment and other anti-VEGF or mTOR targeting agents are also being used [18]. There are currently several other ongoing Phase III clinical trials evaluating the safety and efficacy of combination chemotherapy in the relapsed metastatic RCC such as lenvatinib plus everolimus vs lenvatinib plus pembrolizumab vs sunitinib (NCT02811861), nivolumab plus cabozantinib vs sunitinib (NCT03141177). Studies of sunitinib in combination with nivolumab and of pazopanib in combination with either nivolumab or pembrolizumab were stopped early because of apparent synergistic fatigue and liver toxicity [30].

Non-clear cell RCC

Non-clear cell RCC tumors make up a much rarer population of renal cell cancers and include papillary RCC which makes up about 15–20% of all RCCs and chromophobe RCC, which makes up about 4% of all RCCs [31,32]. Given rarity of these histologic subtypes, treatment consensus has not been well defined, although use of TKIs (bevacizumab with erlotinib, sunitinib, cabozantinib) is generally considered the first-line for patients with advanced or metastatic papillary RCC. Ipilimumab plus nivolumab or single agent nivolumab are both acceptable options, based on data from clear cell RCC studies as well as observational studies [33], though not always widely accepted standard treatment options [34]. For combination ICI in non-clear cell RCC histologic subtypes, patients treated with PD-1- or PD-L1-targeting agent as monotherapy or in combination with another systemic agent, the ORR for papillary RCC was 29%, the overall median time-to-treatment failure for all histologic subtypes was 4 months with the median OS over 1 year with similar results with nivolumab monotherapy [33,35]. Pembrolizumab also

demonstrated efficacy in papillary RCC in an open-label, nonrandomized Phase II KEYNOTE-427 trial with an ORR of 28% and 1-year OS of 74% [36].

Urothelial & bladder carcinoma

The overall survival for patients with metastatic bladder cancer who received standard-of-care platinum-based chemotherapy is estimated to be 9–15 months and is reduced to $\leq$7 months in relapsed patients who received platinum-based chemotherapy [37,38]. For patients with poor performance status and who are ineligible to receive cisplatin due to renal insufficiency, there are a limited number of treatment options available [39].

Nonmuscle-invasive bladder cancer

Nonmuscle-invasive bladder cancer (NMIBC) is the most common presentation of bladder cancer and given high risk for recurrence and progression in those who have high-grade urothelial bladder cancers, Bacillus Calmette-Guérin (BCG) has been used as adjuvant therapy to prevent recurrence. While BCG is generally effective, recurrence still occurs in up to 80% of patients and progression to muscle-invasive bladder cancer (MIBC), which brings excessive morbidity and mortality, still occurs in up to 45% of patients [40]. The use of ICI in the form of pembrolizumab has garnered benefit based on the results of KEYNOTE-057 [41]. The trial aimed to evaluate the ability of pembrolizumab to induce a complete response and prolong disease-free survival among patients with high-risk NMIBC who were unresponsive to BCG and refused or were ineligible for cystectomy. After 3 months of treatment, pembrolizumab produced a complete response in 40.2% of patients with high-risk NMIBC whose disease was no longer responding to BCG. In addition, among the patients (n = 40) who were able to achieve a complete response (CR) at 3 months, majority at 72.5% were able to maintain at a range of 4–26.3 months at a median follow-up of 14 months and 80.2% of pts had a CR duration of exceeding 6 months. This has led to the benefit of pembrolizumab in this population of patients with eventual FDA approval on 8 January 2020 as primary treatment for high-risk NMIBC with carcinoma *in situ* with or without papillary urothelial cancers who are cystectomy ineligible or otherwise refuse to undergo cystectomy.

Muscle-invasive bladder cancer

Radical cystectomy and bilateral pelvic lymphadenectomy remain the standard-of-care for patients with MIBC. Treatment with ICIs is not currently approved for MIBC, though early-phase studies have reported high pathologic CR rate (up to 42%) in with neoadjuvant pembrolizumab and atezolizumab, suggesting a role for ICIs in this setting [42,43]. Results reported recently as part of the Phase III IMvigor010 trial showed no significant disease-free survival benefit with adjuvant atezolizumab compared with observation alone [44]. Multiple trials are ongoing in this space to determine whether additional ICIs in combination with standard-of-care chemotherapy may further improve responses. For instance, KEYNOTE-866/MK-3475-866 (NCT03924856) looks at the utility of perioperative pembrolizumab in addition to neoadjuvant gemcitabine and cisplatin prior to definitive radical cystectomy with the primary end point of pathologic complete response and event-free survival in the overall population as well as those with tumors that express PD-L1 with a combined positive score (CPS) $\geq$10.

Metastatic urothelial cancer

Cisplatin-based combination chemotherapy such as dose-dense methotrexate, vinblastine, doxorubicin, cisplatin (MVAC) is the first-line treatment for metastatic urothelial cancer. For patients who are ineligible for cisplatin-based chemotherapy, carboplatin-based regimen or a nonplatinum combination such as paclitaxel and gemcitabine are options. However, up to 50% of advanced urothelial carcinoma patients are not candidates for cisplatin-based therapy due to age or comorbidities [39] and in recent years, this has been the driving force to explore ICI treatment in this subgroup.

Both anti-PD-1 (pembrolizumab) and anti-PD-L1 agents (atezolizumab) have been approved initially in 2017 by the FDA to treat metastatic urothelial bladder cancer as an alternate first-line treatment. Pembrolizumab demonstrated an ORR of 29% with 7% CR and 22% PR in a single arm Phase II KEYNOTE-052 trial with cisplatin-ineligible patients with advanced urothelial cancer not previously treated with systemic chemotherapy [45]. The median follow-up time was 9.5 months and the ORR was higher in patient with PD-L1 expression $\geq$10%. Atezolizumab was also initially approved as a first-line treatment for advanced urothelial carcinoma for patients ineligible for cisplatin-based chemotherapy based on the single-arm Phase II IMvigor120 trial with 123 cisplatin-ineligible patients with advanced urothelial cancer not previously treated with systemic chemotherapy, where it

Table 2. US FDA-approved second-line immune checkpoint inhibitors for metastatic bladder cancer.

Trial	Phase	Mechanism of action	Dosage	Primary end points	Secondary end points	Results of primary end point
CheckMate-275	Single-arm Phase II	PD-1 inh	Nivolumab 3 mg/kg IV every 2 weeks (n = 265)	ORR	OS	Confirmed objective response = 28.4%, 95% CI: 18.9–39.5
KEYNOTE-045	III	PD-1 inh vs. chemotherapy	Pembrolizumab 200 mg IV every 3 weeks	OS + PFS	ORR	Pembrolizumab OS = 10.3 mos vs chemotherapy = 7.4 mos; p = 0.002
IMvigor210	Single-arm, two-cohort, Phase II trial	PD-L1 inh	Atezolizumab 1200 mg IV every 3 weeks	Independent review and investigator-assessed ORR	AEs and SAEs	ORR for IC2/3: 27%; IC1/2/3: 18%; all patients (15%)
JAVELIN	Ib	PD-L1 inh	Avelumab 10 mg/kg IV every 2 weeks	Safety and tolerability	ORR, PFS, OS	ORR independent central review = 18.2%
Study 1108	I/II	PD-L1 inh	Durvalumab 10 mg/kg every 2 weeks	Safety	Confirmed ORR	AEs: fatigue (13.1%), diarrhea (9.8%) and decreased appetite (8.2%); ORR was 31.0% (95% CI: 17.6–47.1)

AE: Adverse event; AE: Adverse effect; DLT: Dose-limiting toxicity; Inh: Inhibitor; IV: Intravenously; MTD: Maximum tolerated dose; ORR: Objective response rate; OS: Overall survival; PD-L1: PD-1 ligand; PFS: Progression-free survival; q: Every; RD: Recommended dose; SAE: Serious adverse event.

demonstrated an ORR of 23% with 9% CR [46]. At a median follow-up time of 17 months, median duration of response had not been reached. However, the US FDA issued a guidance in 2018 based on the Data Monitoring Committee's recommendation, after it was found that patients with PD-L1-low status receiving monotherapy with atezolizumab (in IMvigor130 trial) and pembrolizumab (in KEYNOTE-361 trial) had decreased survival compared with patients who received cisplatin- or carboplatin-based chemotherapy. The revised US FDA guidance and label for these two ICIs states they can only be offered to patients in the first-line metastatic setting for those who are not eligible for any platinum-containing chemotherapy or not eligible for cisplatin-containing chemotherapy and whose tumors/infiltrating immune cells express a high level of PD-L1 [47]. The IMvigor130 trial has therefore closed the monotherapy arm but continues with the combination arms to determine the effects of atezolizumab in combination with gemcitabine- and platinum-based chemotherapy in patients with treatment-naive metastatic urothelial cancer; preliminary results have shown an increase in PFS (6.3 months in arm with chemotherapy without atezolizumab and 8 months with chemotherapy with atezolizumab) [48]. There was no significant change in OS, which ranged from 13–15 months, and in patients who were treated with single agent atezolizumab, chemotherapy only, or combination therapy, ORRs were 47, 23 and 44%, and CR rates were 13, 6 and 7%, respectively.

Second-line therapy for metastatic urothelial cancer

Five anti-PD-1 and anti-PD-L1 agents have also been approved by the US FDA for second-line treatment of metastatic urothelial carcinoma in patients who have been refractory to platinum-based chemotherapy. These include pembrolizumab, nivolumab, atezolizumab, durvalumab and avelumab (see Table 2).

In a randomized, open-label, Phase III KEYNOTE-045 trial in patients with metastatic urothelial cancer who have progressed during or after platinum-based chemotherapy, pembrolizumab demonstrated a median OS of 10 months compared with single agent chemotherapy (paclitaxel, docetaxel, or vinflunine) with 7% CR and 22% PR. PFS was approximately the same in both groups. Pembrolizumab also had improve response rate (21 vs 11% and a higher rate of duration of response lasting >12 months (68 vs 35%). Adverse events of grade 3 or higher were also less frequent (17% vs 50%) [49]. Median follow-up time was 27 months and both 1- and 2-year OS rates were higher with pembrolizumab (44 and 27%) than chemotherapy (30 and 14%) [50]. These results led to the US FDA approval for pembrolizumab use in second-line therapy and continues to have level 1 evidence for use among all the other agents [51].

Atezolizumab is also approved for patients with metastatic or locally advanced urothelial cancer who have progressed during or after platinum-based chemotherapy based on increase ORR and duration of response. A multicenter Phase II trial investigating the use of atezolizumab in inoperable locally advanced urothelial cancer patients, which showed an ORR of 15% in all patients, with an ORR up to 27% with higher PD-L1 expression [52] which led to the initial US FDA approval of this drug for use as second line therapy after failure from chemotherapy.

The Phase III randomized control trial IMvigor211 investigated the use of atezolizumab (1200 mg IV every 3 weeks) to chemotherapy (investigator's choice of vinflunine, paclitaxel or docetaxel) in patients with metastatic urothelial cancer [53]. However, there was no significant improvement in median OS (11.1 vs 10.6 months, HR: 0.87, 95% CI: 0.63–1.21) and the ORRs between both arms were also similar. The median duration of response was longer atezolizumab compared with chemotherapy (15.9 vs 8.3 months). Nonetheless, the US FDA label was maintained for use as second-line treatment in metastatic urothelial cancers.

Nivolumab demonstrated activity in Phase I and II studies on patients who had progressed on previous platinum-based therapy. In two Phase II studies with median follow-up time ranging from 7 to 15 months, the ORR was noted in 24–28% of patients [54,55], leading to its US FDA approval in February 2017 for second-line treatment of metastatic urothelial carcinoma. Combination therapy with different doses of nivolumab and ipilimumab has been studied in the open-label Phase II CheckMate-032 study, which demonstrated an ORR of 38% with combination therapy with nivolumab (1 mg/kg) and ipilimumab (3 mg/kg) versus 26% with nivolumab alone and 27% with nivolumab (3 mg/kg) and ipilimumab (1 mg/kg) [56]. The median PFS across the arms was 4.9, 2.8 and 2.6 months. Median OS was 15.3, 9.9 and 7.4 months. These data suggested superiority of nivolumab 1 mg/kg plus ipilimumab 3 mg/kg regimen but further investigations are pending, and this regimen has not been US FDA approved.

Based upon the results of two Phase I expansion cohorts, avelumab was approved by the US FDA in May 2017 for treatment of advanced urothelial carcinoma that progressed during or after platinum-based chemotherapy [57]. The ORR in these studies was 17% with 6% CR, 11% PR, and 23% stable disease [58]. The response rate was higher in high PD-L1 expression tumors compared to tumors with low or negative PD-L1 expression. In addition, early press release reports of JAVELIN Bladder 100 trial showed that maintenance avelumab in metastatic urothelial cancer patients who have received upfront gemcitabine and cisplatin or carboplatin who have achieved either stable disease or complete/partial responses improved OS [59].

Durvalumab was also approved by the US FDA in May 2017 for treatment of advanced urothelial carcinoma that has progressed during or after previous platinum-based chemotherapy, either for metastatic disease or for progressive disease less than 12 months after adjuvant or neoadjuvant chemotherapy. This was based on a Phase I/II multicenter open-label study [60,61] which demonstrated an ORR was seen in 18% of patients. The response rate was higher in high PD-L1 expression tumors compared to tumors with low or negative PD-L1 expression.

The optimal sequence of ICIs and/or combinations with chemotherapy and emerging antibody–drug conjugates will be further explored in years to come. There are also several ICIs and combination therapies that are under active investigation (see Table 3). Meanwhile, the failure of atezolizumab in the neoadjuvant setting in unselected patients raises questions about patient selection, especially in the context of the emerging role of pembrolizumab in localized carcinoma *in situ* that has recently been US FDA approved.

Prostate adenocarcinoma

Prostate cancer cells overexpress several highly immunogenic tumor-associated antigens such as tartrate-resistant acid phosphatase, prostate-specific antigen (PSA), alkaline phosphatase, and prostatic acid phosphatase, which have been targets for immunotherapy, specifically vaccines [62,63]. Prostate cancer tissue is marked by a large inflammatory infiltrate of T cells (tumor-infiltrating lymphocytes [TILs]) within the tissue and its microenvironment [64,65]. The presence of TILs has been shown to correlate with prognosis, with lower density of associated with high risk of tumor progression and of a fatal disease [66]. However, prostate cancer cells are not responsive to immunotherapy due to decreased immunogenicity of surface antigens, nonreactive TIL and regulatory T-cell infiltrate, which is thought to result in an inability mount an immune response, thereby leading to evasion of recognition by immune system as well as decreased effectiveness of immune response [67–69].

To date, Sipuleucel-T (Provenge; Dendreon Inc.) is the only approved vaccine therapy for prostate cancer. It was approved by the US FDA in 2010 to treat men with asymptomatic or minimally symptomatic metastatic castration-resistant prostate cancer (mCRPC) [70]. It is a dendritic cell vaccine made from autologous peripheral blood mononuclear cells (PMBCs), which enhances the T cell response to prostatic acid phosphatase. PMBCs are collected from patient via leukapheresis, then pulsed with a fusion protein made from human GM-CSF and prostatic acid phosphatase and re-infused into the patient intravenously after 3 days. Patients undergo this treatment every 2 weeks for a total of three doses [71].

In the original randomized Phase III trial against placebo, sipuleucel-T had increased OS (26 vs 21 months) which was associated with T cell stimulation which was eightfold higher at 8 weeks in sipuleucel-T-treated patients [72]. Time to disease progression was also higher in the sipuleucel-T group (11.7 vs 10 weeks) but was

Table 3. Ongoing trials with immunotherapy agents in muscle-invasive bladder cancer.

ClinicalTrials.gov Identifier:	Immunotherapy and other agents	Name of clinical trial
NCT03832673	Pembrolizumab and Epacadostat	PECULIAR: an open label, monocenter, single-arm, phase II study of neoadjuvant pembrolizumab and epacadostat, preceding radical cystectomy, for patients with muscle-invasive bladder cancer
NCT03674424	Avelumab	Avelumab as neoadjuvant therapy in subjects with urothelial muscle invasive bladder cancers (AURA trial)
NCT03549715	Durvalumab Tremelimumab MVAC	NEoadjuvant dose-dense MVAC In cOmbination with durvalumab and tremelimumab in muscle-invasive urothelial carcinoma
NCT03244384	Pembrolizumab	Testing MK-3475 (pembrolizumab) after surgery for localized muscle-invasive bladder cancer and locally advanced urothelial cancer
NCT03747419	Avelumab	Avelumab and radiation in muscle-invasive bladder cancer
NCT03775265	Atezolizumab MVAC	Phase III randomized trial of concurrent chemoradiotherapy with or without atezolizumab in localized muscle invasive bladder cancer (Study SWOG/NRG 1806)
NCT02621151	Pembrolizumab Gemcitabine	A Phase II trial of MK3475 in combination with gemcitabine and concurrent hypofractionated radiation therapy as bladder sparing treatment for muscle-invasive urothelial cancer of the bladder
NCT03518320	Nivolumab Gemcitabine-Releasing Intravesical System (GemRIS)/TAR-200	A multicenter study of TAR-200 in combination with nivolumab (OPDIVO) in subjects with muscle-invasive urothelial carcinoma of the bladder who are scheduled for radical cystectomy and are ineligible for or refusing platinum-based neoadjuvant chemotherapy
NCT02989584	Atezolizumab Gemcitabine Cisplatin	A pilot safety study and single arm Phase II study of gemcitabine and cisplatin with atezolizumab (MPDL3280A) in Patients With Metastatic and Muscle Invasive Bladder Cancer, respectively
NCT02736266	Pembrolizumab	An open label, single-arm, Phase II study of neoadjuvant pembrolizumab (MK-3475) before cystectomy for patients with muscle-invasive urothelial bladder cancer
NCT02812420	Durvalumab Tremelimumab	A pilot pre-surgical study evaluating anti-PD-L1 antibody (Durvalumab) plus anti-CTLA-4 (Tremelimumab) in patients with muscle-invasive, high-risk urothelial carcinoma who are ineligible for cisplatin-based neoadjuvant chemotherapy
NCT02845323	Nivolumab Urelumab	Randomized Phase II study of neoadjuvant nivolumab with and without urelumab in cisplatin-ineligible or chemotherapy-refusing patients with muscle-invasive urothelial carcinoma of the bladder
NCT03617913	Avelumab MVAC	Phase II study evaluating combination chemotherapy + radiotherapy (RT) with avelumab in muscle invasive bladder cancer
NCT04073160	Durvalumab Tremelimumab	TRIO bladder: a Phase Ib study of durvalumab (MEDI 4736) plus tremelimumab followed by concurrent durvalumab plus bladder radiation, based on molecular subtypes in muscle-invasive bladder cancer

not statistically significant. Although PFS, the primary end point, was not statistically significant, the beneficial effect of sipuleucel-T was further illustrated in another randomized Phase III trial, which demonstrated a 33% decrease in risk of death (hazard ratio of 1.5) when treated with the vaccine [71]. Subsequently, a larger Phase III IMPACT trial was designed with OS as the primary end point. It demonstrated an OS benefit of 4 months (25.8 vs 21.7 months in placebo group) with a similar time to disease progression but an increase in immunogenic response as measured by antibody titers against prostate cancer antigens PA2024 and prostatic acid phosphatase [73]. Sipuleucel-T administration was associated with nonspecific constitutional symptoms in comparison to the placebo and was generally well-tolerated [74].

ICI therapy

Unfortunately, unlike RCC and bladder cancer the role of ICIs in prostate cancer is limited to small subpopulations. Response to ICI therapy is driven by microsatellite instability and several reports have investigated the incidence of deficient mismatch repair (dMMR) in prostate cancer, which has ranged from 2 to 4% [6,75,76]. Ipilimumab, given at dose of 10 mg/kg every 3 weeks for four cycles has been tested in men with mCRPC in two Phase III clinical trials but has failed to show any improvement in OS [77,78]. The OS was 11.2 vs 10 months in the placebo group with a HR of 0.85 when pretreated with docetaxel [77] or chemotherapy-naive patients [78], but had some increase in PFS (5.6 vs 3.8 months) and PSA response (23 vs 8%) in chemotherapy-naive patients.

The prostate adenocarcinoma cohort (n = 23) of the non-randomized Phase Ib KEYNOTE-028 trial suggested that treatment with pembrolizumab results in durable responses in mCRPC patients with PD-L1 expression $\geq$1% with an ORR of 17% and median duration of response of 13.5 months [79]. The KEYNOTE-199 Phase II trial examined the effect of pembrolizumab in men with chemotherapy-refractory metastatic prostate cancer demonstrated a disease control rate (defined as the percentage of patients with a confirmed radiographic objective response of any duration or stable disease, or a non-complete response or nonprogressive disease for 6 months or longer)

of about 10% at 6 months regardless of PD-L1 expression and a median duration of response of approximately 17 months [80]. However, the disease control rate was higher (22%) in patients with bone-predominant disease, suggesting antitumor activity in this subgroup of patients.

Ongoing studies are evaluating ICI combinations with ICI like durvalumab and PARP-inhibition to determine if synergies exist between standard agents and immunotherapy in prostate cancer [81]. Early studies suggest efficacy of pembrolizumab in patients with mCRPC evidence of progression on enzalutamide [82]. In addition, multiple combination studies including enzalutamide with or without pembrolizumab in mCRPC (KEYNOTE-641; NCT03834493 or first-line enzalutamide with or without pembrolizumab in the mCSPC (KEYNOTE-991; NCT04191096) are currently enrolling.

Testicular cancer

Testes belong to a group of immunologically protected sites, with a naturally suppressed immune system that promotes the growth of spermatids. Germ cell tumors (GCT) are divided into pure seminoma and nonseminomatous germ cell tumors (NSGCT), both of which are primary treated with resection. The treatment of relapsed GCT depends on response to prior therapy, location and timing of relapse and tumor histology, and chemotherapy remains the backbone for relapsed disease. PD-1 is poorly expressed in GCT [83] but there is high expression of PD-L1 in seminomas (73–76%) and non-seminomas (64–89%) as detected by IHC, suggesting that treatment with ICI therapy could be promising for GCT [84,85]. Anti-PD-1 agents (nivolumab and pembrolizumab) were tested in a relatively small study comprising of seven patients with platinum-refractory metastatic GCT, which demonstrated an OS ranging from 0.3–20.2 months, with four patients who died after receiving 1 dose of anti-PD-1 agent due to PD [86]. Another small single-institution study examined the effects of pembrolizumab administration on four patients with platinum-refractory GCT and this study demonstrated progression on pembrolizumab [87]. Given these findings, ICI is not currently recommended for GCT and prospective trials in chemotherapy-refractory GCT are warranted.

Conclusion & future direction

Much like the broader field of medical oncology, the dawn of the modern immunotherapy age has created hope and treatment options for patients with advanced disease. Nonetheless, despite these remarkable developments, response rates are not universal and tumor types such as prostate cancer remain largely unimpacted. The next steps in the immunotherapy age will likely seek refinement of the data gathered so far, such as what biomarkers can be used to select patients that are most likely to respond. Also, biomarkers may inform which subpopulations require immunologic escalation with an agent like ipilimumab, which is associated with both responses and life-impacting toxicity. While important in assisting with patient selection, biomarkers may not be the answer for the majority of patients who still do not benefit from immunotherapy. One strategy that may unlock the therapeutic benefits of immunotherapy for a broader population may be derived from the first immunotherapies used in RCC. Cytokines, while toxic, likely impacted more cellular subpopulations in the tumor microenvironment than just T cells. Indeed, emerging data is highlighting that other cell types such as tumor associated macrophages, natural killer cells and myeloid derived suppressor cells may be important players within the tumor immune microenvironment [88,89]. Modern therapeutic technology is refining cytokines and developing immunocytokines that can localize to the tumor microenvironment while sparing systemic toxicities, and these agents may impact the many pleotropic aspects of the tumor microenvironment beyond T cells. These and other strategies may enhance the therapeutic benefits of immunotherapy in genitourinary malignancies and beyond.

Executive summary

- Immunotherapy has revolutionized treatment for several genitourinary cancers.
- Immune checkpoint inhibitors has become a standard second-line therapy for metastatic urothelial cancers that have failed prior chemotherapy.
- Immune checkpoint inhibitors have become standard first-line therapy for metastatic renal cell cancers.
- While vaccine therapy with Sipuleucel-T is considered standard of care for patients with metastatic castration-resistant prostate cancer, the role of immunotherapy in prostate cancer continues to evolve.
- Role of pembrolizumab in metastatic castration-resistant prostate cancer is limited in those with microsatellite instability–high or mismatch repair deficient (dMMR) gene mutations.

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J Aragon-Ching serves on the Speakers' Bureau for BMS, Astellas/Seattle Genetics and serves on the Advisory Board for EMD Serono. SU Gandhy and RA Madan have no conflicts to declare. The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.

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