Checkpoint inhibitors in the treatment of urological malignancies

Lazar S Popovic,1,2 Gorana Matovina-Brko,1,2 Maja Popovic1,2

ABSTRACT

Checkpoint inhibitors are monoclonal antibodies against several different receptors on T-cells or tumour cells expressing receptors for cytotoxic T-lymphocyte antigen 4 (CTLA-4), programmed death-1 (PD-1) and their ligand (PD-L1). Since 2010, numerous trials on different tumour types have been conducted, which was resulted in these drugs being approved for the treatment of melanoma, lung cancer, Hodgkin’s lymphoma and head and neck cancers. Urological cancers, especially urothelial and renal-cell carcinomas, are immunogenic tumours. Since the late 70s, the bacillus Calmette-Gurin (BCG) vaccine has been used for intravesical instillation in non-muscle invasive bladder cancer from the mid-90s up until the discovery of tyrosine kinase inhibitors (TKIs) in 2007, interleukin-2 (IL-2) and interferon alpha (IFNα), which were the standard of care for metastatic renal-cell cancer. Two checkpoint inhibitors are already approved by the Food and Drug Administration: atezolizumab for metastatic urothelial cancer and nivolumab for metastatic renal-cell carcinoma. There are many drugs are in different phases of clinical development. Here we review the current status of checkpoint inhibitors in the treatment of urological tumours.

INTRODUCTION

For a number of years, urological cancers have been considered to be tumours that respond well to immunotherapy. The first immune drug approved by the Food and Drug Administration (FDA) was the bacillus Calmette-Gurin (BCG) vaccine, used for intravesical instillation in non-muscle invasive bladder cancer.1 Since the mid-90s up until the discovery of tyrosine kinase inhibitors (TKIs) in 2007, interleukin-2 (IL-2) and interferon alpha (IFNα), alone or combined, had the overall response rate between 14% and 25%, with the median overall survival (OS) of about 13 months and progression free survival (PFS) of 4 months.2,3 Furthermore, in a meta-analysis, the IFNα was associated with a benefit in the OS relative to different comparators.4 Besides the limited efficacy, the main problem of these therapies was toxicity.2,3,5,6

Recently, sipuleucel-T—a complex treatment for castration-resistant prostate cancer (CRPC)—was approved by the FDA after the confirmed OS benefit in asymptomatic or minimally symptomatic patients.7 Checkpoint inhibitors are monoclonal antibodies against several different receptors on T-cells or tumour cells: cytotoxic T-lymphocyte antigen 4 (CTLA-4), programmed death-1 (PD-1) and their ligand (PD-L1). Since 2010, numerous trials on different tumour types have been conducted and have resulted in the approval of these drugs for the treatment of melanoma,8-10 lung cancer,11-13 Hodgkin’s lymphoma14 and head and neck cancers.15 In urological tumours, nivolumab has been approved for the treatment of metastatic renal cancer (mRCC) after progression on TKI.16 Atezolizumab has been approved in the USA for metastatic urothelial cancer after progression to cisplatin.17

MATERIAL AND METHODS

We conducted a PubMed search with keywords: urothelial cancer immunotherapy, renal cell cancer immunotherapy, prostate cancer immunotherapy, and also reviewed the data from relevant meetings (ESMO, ASCO, ASCO GU) from year 2011 to 2016. Only articles in English were considered.

CHECKPOINT INHIBITORS MECHANISM OF ACTION

Tumour cells produce numerous foreign antigens in the host immune system. Similar to the infectious antigens, the antigen presenting cells (APCs) are responsible for the recognition of these tumour antigens. After determining the foreign antigen, the APC migrate to lymphoid organs, where they introduce the foreign antigen to T-cells. This process requires the activation of the major histocompatibility complex and the T-cell receptor as well as of other costimulatory mechanisms. One of the most important costimulatory mechanisms includes the bond between the CD80 and CD86 receptors, which are expressed on mature APC and which stimulate cytotoxic T-cells to eliminate foreign antigens when attached to CD28.18,19 However, when attached to the CD80 and CD86, the CTLA-4 produces an inhibition signal,
resulting in the absence of T-cell activation. This mechanism is established in order to prevent an uncontrolled activation of the T-cells and consequent autoimmune reactions. In experiments performed on mice, the mice without CTLA-4 have experienced rapid death due to inadequate lymphoproliferation and an excessive autoimmune reaction. 

However, this mechanism also prevents the activation of T-cells against tumour cells and protects the tumour from the immune cell recognition. 

Ipilimumab and tremelimumab are checkpoint inhibitors that bind to the CTLA-4 receptor and prevent it from being connected to the CD80 and CD86. These drugs allow the binding of the CD28 to the above-mentioned receptors and the T-cell activation. In 2011, ipilimumab was approved for the treatment of metastatic melanoma. 

The second significant inhibitory signal to cytotoxic cells is the connection between the PD-1 receptors and its ligands PD-L1 and PD-L2. The PD-1 receptor is situated on T-cells, whereas the ligands can be found both on the immune system cells and on cells of other organs, such as striated muscles, lungs and so on. The PD-L1/2 are also expressed on tumour cells and tumour infiltrate cells and enable the tumour to protect itself from the host immune system. 

Several checkpoint inhibitors can block the bond between the PD-1 and its ligands by connecting to either the PD-1 or PD-L1. Anti-PD-1 agents in urological malignancies are: nivolumab, which is approved for the treatment of mRCC, and pembrolizumab, which is being investigated in the treatment of metastatic urothelial carcinoma (mUC), non-muscle-invading urothelial cancer (UC), prostate cancer and mRCC. Anti-PD-L1 inhibitors are atezolizumab, approved for the treatment of mUC, as well as durvalumab and avelumab, which are currently in the process of being studied in mUC.

**UROTHELIAL CANCER**

In about 70% of the patients with urothelial cancer, the initial presentation is as a non-muscle invasive disease. These patients are treated with repeated transurethral resections, with or without intravesical instillation of chemotherapy or BCG. Although the BCG has minimal benefit in recurrence reduction, these patients often experience non-muscle invasive or muscle invasive relapse. 

In muscle-invasive urothelial cancer, the standard of care is radical cystectomy with previously applied neoadjuvant cisplatinum-based chemotherapy. Treatment of locally advanced metastatic cancers of the urinary bladder is also based on cisplatinum-based chemotherapy. The median OS of patients treated with gemcitabine and cisplatinum is about 14 months. However, 30%–50% of these patients are unfit for cisplatinum treatment, making gemcitabine and carboplatinum the optimal therapeutic option with a 9-month median OS. The only drug in the second line of the treatment approved in Europe is vinflunine, with the median OS of 6 months.

Atezolizumab is a humanised anti-PD-L1 monoclonal antibody. In a phase I trial, atezolizumab was tested in the dose of 1200mg or 15mg/kg, every 3 weeks, in patients with mUC, mRCC, melanoma and lung cancer. Updated results in patients with mUC were reported at ASCO 2015. Out of the 85 previously treated patients enrolled in the study, 46 were immunohistochemically 2/3 (IC2/3) positive to PD-L1, while 38 were IC0/1. Visceral metastasis were present in 77% of the patients. The overall response rate (RR) was 46% for IC2/3 patients; six had a complete remission (CR) and 15 had a partial remission (PR). In patients with IC0/1, the ORR was 16%. A 6-month survival in patients with IC2/3 and IC0/1 was 85% and 71%, respectively. Adverse events (AEs) associated with the study drug were noted in 67% of patients, and grade 3 and 4 AEs in 8%. The most frequent AEs were fatigue, asthenia and nausea.

The phase II study of atezolizumab was divided into two cohorts. The results of cohort 2 were published earlier. Cohort 2 included patients who progressed after ciplatinum-based treatment. The study recruited 310 patients who were treated with atezolizumab 1200mg every 3 weeks. The median age was 66. Groups were stratified according to the PD-L1 expression on immune cells of the tumour infiltrate: IC0 for expression <1%, IC1 1%–5% and IC2/3 >5%. The overall response in the study was 15%; for IC2/3 and IC1/2/3, it was 26% and 18%, respectively. After the median follow-up of 11.7 months, the median survival for IC2/3, IC1, IC0 was 11.4, 6.7 and 6.4 months, respectively. Immune-related AEs of grades 3 and 4 were reported in 5% of the patients, with pneumonitis, transaminitis, rash and dyspnoea being the most common ones. The conclusion of this study was that the overall response rate (ORR) and OS were longer when compared with historical controls, in addition to a very good safety profile.

Based on the results of this study, FDA approved atezolizumab in the treatment of mUC patients who progressed on cisplatin. Cohort 1 included chemo-naïve metastatic patients unfit for cisplatinum-based chemotherapy, which was defined as the glomerular filtration rate >30 and <60 mL/min, Eastern Cooperative Oncology Group (ECOG 2), hearing loss and/or peripheral neuropathy grade ≥2. One hundred and nineteen patients were treated. The median age was 73. Eighteen per cent of patients had previously received (neo)adjuvant treatment, while 10% had been treated with radiotherapy. The overall response rate was 19%, and a response was reported in all the PD-L1 expression groups. Out of 23 patients who responded to the treatment, 22 patients had no progression after the median follow-up of 8.5 months (0.2–14.3). The median OS was 10.6 months. Grades 3 and 4 AEs that occurred in more than 10% of the patients were pruritus, diarrhoea and fatigue, and one patient died of sepsis.

Two phase III studies, based on the results of the phase II study, are ongoing. IMvigor 211 is comparing atezolizumab versus the investigators’ choice (vinflunine, docetaxel and paclitaxel) in patients with mUC who progressed after treatment with a cisplatinum-based
Pembrolizumab is a monoclonal antibody against PD-1 receptors. In phase Ib of the clinical trial KEYNOTE-012, 33 patients with previously treated urothelial cancer were enrolled. The patients were given pembrolizumab 10 mg/kg every 2 weeks, until the disease progression, toxicity or the achievement of the CR. Patients were enrolled in the study if the PD-L1 expression was ≥1%. The median age was 70 years. Thirty-three per cent of the patients had previously received two or three lines of treatment. The ORR was 25% (11% CR, 14% PR), 1 year PFS was 19% and the median survival was 12.7 months. A total of 15% of the patients had AE of grades 3 and 4.36

In the recently published phase III trial (KEYNOTE-045), patients who had previously been treated with one or two lines of the platinum-based protocol were randomised for receiving a therapy of pembrolizumab 200 mg intravenously, every 3 weeks, and the investigators’ choice (docetaxel, paclitaxel and vinflunine). The study included 5,421 patients, and the median age was 67 and 65 years in pembrolizumab and chemotherapy groups, respectively. ORR was 21.1% in the group treated with pembrolizumab and 11.4% in the group which received chemotherapy, while the CR rate was 7% and 3% in pembrolizumab and chemotherapy groups, respectively. PFS did not differ between the groups (HR 0.98; 95% CI 0.81 to 1.19; p = 0.42), while the OS was longer in the group treated with pembrolizumab (10.3 vs 7.4 months, HR 0.73, 95% CI 0.59 to 0.91; p = 0.0022). It is interesting that the patients with the PD-L1 expression of over 10% (tumour and immune cell PD-L1 expression, combined positive score (CPS)) did not experience longer survival in the pembrolizumab group when compared with the general study population. However, the anti–PD-1 antibodies’ benefit was sustained in comparison with chemotherapy (OS 8.0 vs 5.2 months; HR = 0.57; 95% CI 0.37 to 0.88; p = 0.0048). AEs were more common in the chemotherapy group (60.9% vs 90.2%), including the adverse effects of grades 3–5 (15.0% vs 49.4%). Immune-related AEs including thyroid gland abnormalities, colitis and pneumonitis were more frequent in the group treated with pembrolizumab.37

Besides in platinum-pretreated patients, pembrolizumab is also being investigated in cisplatin ineligible patients with mUC. The results of first 100 of planned 374 patients, in phase II study KEYNOTE-052, have been reported. The median age was 75 years, with 13% of patients having undergone perioperative chemotherapy. The majority of patients had visceral metastases, 46% of the patients were ECOG 2/3. The overall RR was 24%, with 6% of CR. In the population with CPS ≥1%, the ORR was 25.4%, CR 6.3% while in those with CPS ≥10%, the ORR was 36.7% and CR 13.3%. The duration of response ≥6 months was achieved in 83% of patients. The most frequent AE was fatigue, which occurred in 14% of cases.38

Apart from the above-mentioned studies, the currently ongoing phase III study (KEYNOTE-341) is focusing on the first-line treatment and assessing pembrolizumab alone or in combination with gemcitabine/cisplatin (or carboplatin) protocol versus gemcitabine/cisplatin (or carboplatin) chemotherapy.39 Pembrolizumab is also being researched in muscle-invasive bladder cancer patients, relapsing after the BCG treatment (KEYNOTE-057).40

Durvalumab is a human anti-PD-L1 antibody assessed in phase I/II trials in patients with advanced urothelial cancer. The difference between the studies involving durvalumab and those of atezolizumab and pembrolizumab is the cut-off value for PD-L1 positivity ≥25% CPS. The study included 61 patients: 40 PD-L1 positive, 21 negative. In 93.4% of cases, the patients had previously received at least one systemic treatment. The primary endpoint was safety, while the secondary endpoint was an objective response. There were no instances of grade 4 and 5 toxicities, while grade 3 toxicities were reported in 4.9% of the patients. The objective RR was 31%; 46.6% in the PD-L1 positive, 0% in the PD-L1 negative. Even though there were no objective responses in the PD-L1 negative patients, following the 12 weeks of treatment the disease control was achieved in 28.6% of patients, compared with 57.1% in the PD-L1-positive patients.41

In the phase I trial (CheckMate-032), nivolumab was applied in 3mg/kg doses every 2 weeks, in patients with mUC, regardless of the PD-L1 status. The primary endpoint was the ORR. A total of 78 patients were treated; two-thirds had previously received more than two lines of systemic therapy. The ORR was 24.4%, median PFS 28 months, while the survival at 12 months was 51.6%. After the median follow-up of 213 days (22–499), one-third of the patients were still receiving the treatment. Two patients died of treatment-related complications (pneumonitis and thrombocytopenia). There were 20.5% of treatment-related AEs.42

In the phase II trial (CheckMate-275),43 265 patients were treated with nivolumab. The patients were stratified according to the PD-L1 expression into groups ≤1% and ≥1% and ≤5% and ≥5%, as well as according to the urothelial cancer subtype (by The Cancer Genome Atlas).44 The objective response rate was 19.9%. In patients with the low PD-L1 expression, the objective RR was 16.1%. The median PFS was 2 months, while the median OS was 8.74 months. Grade 3 and 4 AEs were reported in 18% of cases, the most frequent being diarrhea and fatigue. In 1% death occurred as a consequence of treatment-related complications.45

Avelumab, a humanised anti-PD-L1 antibody was also assessed in solid tumours in the phase Ib study (JAVELIN). The cohort included 44 patients with mUC, previously treated with two lines of treatment on average (median 2, range 1–6). Avelumab was administered in 10 mg/kg doses every 2 weeks. The objective RR was 18.6%, with 2 CRs and 4 PRs, while stable disease (SD) was reported in...
In the PD-L1-positive patients, with the cut-off value of $\geq 5\%$ tumour cells (tumour cell staining (TCS)), the ORR was 50% in comparison with 4.3% in patients with PD-L1-negative tumours. After 6 months, the PFS was 58.3% in the PD-L1-positive group and 16.6% in the PD-L1-negative group, and the 1-year survival was 50.9% (table 1).

The results obtained in the study of durvalumab and avelumab have led to the design of the phase III trials. Durvalumab is being investigated in the DANUBE trial, alone or in the combination with tremelimumab, versus the cisplatin-based protocol. Avelumab is being investigated in the JAVELIN 100 trial as a maintenance therapy versus placebo, in patients who achieved at least the CR after having undergone a treatment with a cisplatin-based protocol.

### Table 1 Efficacy of checkpoint inhibitors in the treatment of locally advanced/metastatic urothelial carcinoma

<table>
<thead>
<tr>
<th>Agent/study</th>
<th>Study phase</th>
<th>Indication</th>
<th>Number of patients</th>
<th>Results</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Atezolizumab (PCD4989g)</td>
<td>Phase I</td>
<td>Platinum-pretreated</td>
<td>85</td>
<td>ORR 46% (IC2/3), 16% (IC0/1); 6 months OS 85% (IC2/3); 71% (IC0/1)</td>
<td>31</td>
</tr>
<tr>
<td>Atezolizumab (IMvigor210, Cohort 2)</td>
<td>Phase II</td>
<td>Platinum-pretreated</td>
<td>310</td>
<td>ORR 15% IC2/3 ORR 26%, medOS 11.4 months; IC1 6.7 months; IC0 6.4 months</td>
<td>17</td>
</tr>
<tr>
<td>Atezolizumab (IMvigor210, Cohort 1)</td>
<td>Phase II</td>
<td>Platinum-pretreated</td>
<td>119</td>
<td>ORR 19%; medOS 10.6 months</td>
<td>32</td>
</tr>
<tr>
<td>Pembrolizumab (KEYNOTE-012)</td>
<td>Phase Ib</td>
<td>Platinum-pretreated</td>
<td>33</td>
<td>ORR 25% (11% CR, 14% PR); medOS 12.7 months</td>
<td>36</td>
</tr>
<tr>
<td>Pembrolizumab versus investigators choice (KEYNOTE-045)</td>
<td>Phase III</td>
<td>Platinum-pretreated</td>
<td>542</td>
<td>ORR Pembro versus IC: 21.1% versus 11.4%; CR 7% versus 3.3%; medOS: 10.7 versus 7.4 months (HR 0.73, $p=0.0022$)</td>
<td>37</td>
</tr>
<tr>
<td>Pembrolizumab (KEYNOTE-052)</td>
<td>Phase II</td>
<td>Platinum-pretreated</td>
<td>100</td>
<td>ORR 24%, CR 6%; PD-L1 CPS $\geq$1% ORR 25.4% CR 6.3%; PD-L1 CPS $\geq$10% ORR 36.7% CR 13.3%; DOR $\geq$6 months 83%</td>
<td>38</td>
</tr>
<tr>
<td>Durvalumab</td>
<td>Phase I/II</td>
<td>Pretreated</td>
<td>61</td>
<td>ORR 31%; PD-L1 CPS $\geq$25%; ORR 46.4% 3 months, DCR 57.1%; PD-L1 CPS &lt;25%; ORR 0% 3 months, DCR 28.6%</td>
<td>41</td>
</tr>
<tr>
<td>Nivolumab (CheckMate-032)</td>
<td>Phase I</td>
<td>Platinum-pretreated</td>
<td>79</td>
<td>ORR 24.4%, 1-year OS 51.3%</td>
<td>42</td>
</tr>
<tr>
<td>Nivolumab (CheckMate-275)</td>
<td>Phase II</td>
<td>Platinum-pretreated</td>
<td>265</td>
<td>ORR 19.9%; mOS 8.7 months</td>
<td>43</td>
</tr>
<tr>
<td>Avelumab (JAVELIN)</td>
<td>Phase I</td>
<td>Platinum-pretreated</td>
<td>44</td>
<td>ORR 18.6%, 2 pt CR, 4pt PR; PD-L1+ (cut-off 5%): ORR 50%; PD-L1+: 16.6%; 1 year OS 50.9%</td>
<td>45</td>
</tr>
</tbody>
</table>

CPS, combined positive score; CR, complete remission; DCR, disease control rate; IC (tumour infiltrating), immune cell; medOS, median overall survival; ORR, overall response rate; OS, overall survival; PD-L1, programmed death ligand 1; PR, partial remission.

RENALED CARCINOMA

Renal cell carcinoma is an immunogenic tumour. In RCC tumour infiltrate consists of a great number of T-cells, natural killer (NK) cells, dendritic cells (DCs) and macrophages. As a consequence, the immunotherapy with IL-2 and INFα was the recommended choice for the treatment of mRCC from mid-1990 until 2007, when results of the initial trails with anti-vascular endothelial growth factor (VEGF) drugs led to a change of the standards.

Another attempt of introducing immunotherapy was trial with first checkpoint inhibitor ipilimumab. In the phase II study, 61 patients with mRCC were treated in two cohorts: in cohort A, the loading dose of 3 mg/kg was followed by further therapies of 1 mg/kg, while in cohort B, the administration of the 3 mg/kg dose was continued. The objective RR was higher in the cohort B: 12.5% versus 5%. The most common AEs were enteritis and endocrinopathies (hypophysitis and adrenal insufficiency). However, the patients who had grade 3 and 4 AEs were not included in the analysis.

17 patients. In the PD-L1-positive patients, with the cut-off value of $\geq 5\%$ tumour cells (tumour cell staining (TCS)), the ORR was 50% in comparison with 4.3% in patients with PD-L1-negative tumours. After 6 months, the PFS was 58.3% in the PD-L1-positive group and 16.6% in the PD-L1-negative group, and the 1-year survival was 50.9% (table 1).
toxicities had a treatment response of 30%, versus 0% in patients who experienced no toxicities.50

The phase I and II trials that assessed nivolumab in different solid tumours included patients with RCC. In the dose-escalation phase II trial, patients with mRCC achieved PR.24 In the extended analysis of 296 patients with melanoma, non-small cell lung cancer, CRPC, RCC and colorectal cancer, nivolumab was given in doses ranging from 0.1 up to 10mg/kg, every 2 weeks. The response was achieved in 9 of 33 patients (27%).32

In the phase II trial, patients were randomised into three cohorts depending on the nivolumab dosage: 0.3mg/kg (A), 2mg/kg (B) and 10mg/kg (C). Seventy per cent of the patients had been previously treated with more than one line of treatment. The ORR was 20%, 22% and 20%, the median OS 18.2, 25.5 and 24.7 months, while the median PFS was 2.7, 4.0 and 4.2 months in cohorts A, B and C, respectively. AEs of grade 3 and 4 occurred in 11% of the patients.51

Notable results of nivolumab in the early stage trials led to the phase III trial (CheckMate-025), where nivolumab was assessed in a 3mg/kg dosage every 2 weeks, versus everolimus 10mg/once daily. The study included 821 patients, and the primary endpoint was OS, with the secondary being ORR and safety. The median OS was 25 months in the group treated with nivolumab versus 19.6 months achieved in the group treated with everolimus (HR=0.73, 95% CI 0.57 to 0.93; p=0.002). The ORR was 25% versus 5% (OR, 5.98; 95% CI 3.68 to 9.72; p<0.001) in the group treated with nivolumab and everolimus, respectively. The PFS did not differ (4.6 vs 4.4 months; HR=0.88; 95% CI 0.75 to 1.03; p=0.11). Nivolumab proved to be less toxic, since AEs of grade 3 and 4 were more common in the everolimus treatment group (37% vs 19%). The patients were also stratified in accordance with the PD-L1 TCS, with the cut-off value of ≥1%. The PD-L1-positive patients had worse survival in both cohorts, when compared with the PD-L1-negative patients: nivolumab 21.8 versus everolimus 18.8 months in the PD-L1+, compared with nivolumab 27.4 versus everolimus 21.2 in the PD-L1− group. Nivolumab showed benefits in both groups of patients.16 The subsequent subanalysis demonstrated the survival benefits of nivolumab in each subgroup, including the Karnofsky Performance Status 90−100 versus 90, Heng risk groups and previous treatment with sunitinib, pazopanib or IL-2.24

Anti-PD-L1 antibodies were investigated in the phase I trials in patients with mRCC, BMS-936559 is a human IgG4 antibody which in phase I trial, 2 out of 17 patients achieved a response that lasted 4 and 17 months.55

In the phase I study, atezolizumab was investigated in a dose escalating study whereby it was administered in doses between 3 and 20mg/kg. Most of the patients (83%) had been heavily pretreated. A total of 53 patients were included, and the 6 month PFS was 50%. Treatment-related AEs of grade 3 and 4 were observed in 13% of the patients.60 In the extended cohort (RCC patients from the PCD4989g trial), 70 patients were tested, out of which 63 with clear-cell histology. The patients had been previously treated with TKI (63%), bevacizumab (21%) and mTOR inhibitor (34%). The primary endpoint was safety, while the secondary endpoints were the ORR and immune-related AEs. Patients were stratified according to the IC score into IC1/2/3 (PD-L1 positive ≥1% immune cell infiltrate) and IC0 patients. There were no grade 4 and 5 AEs in the study. Drug-related AEs of grade 3 were reported in 17% of the patients, while immune-related AEs occurred in 4% of the patients. The response was reported in 15% of the patients; 18% in PD-L1+ and 9% in PD-L1− patients. The response was also reported in in 22% of patients with non-clear-cell histology. The OS was 28.9 months, while the PFS was 5.6 months. The 1-year PFS was 81% and showed no difference with regards to the PD-L1 IC status, while the 2-year survival was longer in the PD-L1+ group: 85% versus 51%53 (table 2).

### Table 2  Efficacy of checkpoint inhibitors in the treatment of mRCC

<table>
<thead>
<tr>
<th>Agent/study</th>
<th>Study phase</th>
<th>Indication</th>
<th>Number of patients</th>
<th>Results</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ipilimumab</td>
<td>Phase II</td>
<td>R/R mRCC</td>
<td>61</td>
<td>1mg/kg: ORR 5%; 3mg/kg: ORR 12.5%</td>
<td>50</td>
</tr>
<tr>
<td>Nivolumab</td>
<td>Phase I/II</td>
<td>R/R mRCC</td>
<td>33</td>
<td>ORR 27%</td>
<td>52</td>
</tr>
<tr>
<td>Nivolumab</td>
<td>Phase II</td>
<td>R/R mRCC</td>
<td>168</td>
<td>ORR: cohort A 20%; cohort B 22%;</td>
<td>53</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>cohort C 20%, medOS: A 18.2 months, B 25.5 months, C 24.7 months</td>
<td></td>
</tr>
<tr>
<td>Nivolumab versus everolimus</td>
<td>Phase III</td>
<td>R/R mRCC</td>
<td>821</td>
<td>MedOS: Nivo 25 months versus. Eve</td>
<td>16</td>
</tr>
<tr>
<td>(CheckMate-025)</td>
<td></td>
<td></td>
<td></td>
<td>19.6 (HR=0.73, p=0.002)</td>
<td></td>
</tr>
<tr>
<td>Atezolizumab (PCD4989g)</td>
<td>Phase I</td>
<td>R/R mRCC</td>
<td>70</td>
<td>ORR 15%; IC1/2/3 18%, IC0 9%;</td>
<td>57</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>medOS 28.9 months; medPFS 5.6 months</td>
<td></td>
</tr>
</tbody>
</table>

CR, complete remission; IC (tumour infiltrating), immune cell; medPFS, median progression-free survival; mRCC, metastatic renal cell carcinoma; ORR, overall response rate; OS, overall survival; PR, partial remission.
In the treatment of UC immunotherapy combinations were introduced only in the phase III trials.31 On the other side, several early phase trials with mRCC, assessing combinations of checkpoint inhibitors with other agents, are currently being carried out. Preclinical models have demonstrated that VEGF inhibitors can antagonise immunosuppression and form potent antitumour T-cells. Tumour cells secrete VEGF-A and VEGF signalling decreases DCs costimulatory molecule expression and T-cell priming, and also encourages the formation of myeloid-derived suppressor cells. Sunitinib (and bevacizumab) reverse those effects. Sunitinib also blocks the signal transducer and activator of transcription 3 (STAT3). The decrease of STAT3 signalling diminishes formation of Treg cells and promotes the formation of T-helper cells secreting IFNγ.58,59 These data present the basis for combining these agents and immunotherapy. The anti-CTLA4 antibody tremelimumab was studied in combination with sunitinib in a dose-escalating phase I trial. Due to a high incidence of acute renal failure, this combination was not further investigated.60 Nivolumab was assessed in the phase I trial in combination with sunitinib or pazopanib. Patients had previously received at least one line of therapy for mRCC. Sunitinib (S) and pazopanib (P) were administered in standard doses, while nivolumab was given in a dose-escalating manner, initially 2mg/kg (N2), followed by 5mg/kg (N5). In combinations with sunitinib, there was no dose-limiting toxicities (DLTs) in N2 and N5 groups after seven patients had been enrolled in each group. In P N2 group, there were four DLTs presented as elevation of AST/ALT and fatigue, which resulted in the closure of the cohort. Nineteen more patients were additionally included in the S N5 group. Grade 3 and 4 AEs were reported in 73% of the patients treated with sunitinib and in 60% of those treated with pazopanib. The ORR was 52% in the S and 45% in the P group.61

Nivolumab was also assessed in combination with ipilimumab in the phase I trial. Patients were randomised into a group receiving nivolumab 3mg/kg and ipilimumab 1mg/kg (N3I1) and a group treated with four cycles of nivolumab 1mg/kg and ipilimumab 3mg/kg, followed by nivolumab 3mg/kg until the progression/toxicity (N3I3). Forty-four patients were enrolled in the study, out of which 77% had been previously treated with systemic therapy. Grade 3 and 4 toxicities occurred in 19 patients (43%); 5 in the N3I1 and 14 in the N1I3 group. The objective response rate was 29% in the N3I1% and 39% in the N1I3 group, with 33% and 39% of SD in both groups, respectively. Time to progression was approximately the same in both groups. Based on the results of this study, the phase III trial, in which the nivolumab and ipilimumab combination was compared with the standard sunitinib treatment, has been designed.62 63

PROSTATE CANCER

Ipilimumab was assessed in early phase trials in prostate cancer, alone or in combination with other drugs or radiotherapy.64-66 In the phase III trial, ipilimumab was assessed versus placebo after palliative 8Gy irradiation of bone metastases. Seven hundred and ninety-nine patients who had progressed after the docetaxel treatment were enrolled. The PSA decline was reported in 13.1% of the patients treated with ipilimumab and in 5.3% of the patients on placebo. The OS was 11.2 months on ipilimumab and 10 months on placebo, which was not statistically significant. In the post hoc analysis, the patients with good prognostic factors (no visceral metastases, haemoglobin level over 11.0mg/dL, alkaline phosphatase less than 1.5 upper limit normal) experienced a benefit of the ipilimumab treatment. The OS was 22.7, versus 15.8 months in the placebo group (HR 0.62; 95% CI 0.45 to 0.86; p=0.0038).67

Tremelimumab was examined together with bicalutamide following the biochemical progression without a radiological confirmation of the disease, after primary surgical treatment and/or radiotherapy. Eleven patients were enrolled, and the primary endpoint was safety. The most frequent AEs of grade 3 and 4 were diarrhoea and rash.68

Pembrolizumab was investigated in mCRPC in the phase II trial in combination with enzalutamide. Twenty patients were enrolled, and the primary endpoint was the PSA <50% decline. In four out of 20 patients, serum PSA dropped below 0.01ng/mL, and these patients have been in continuous remission for 16-61 months. Seven patients had disease stabilisation for 9-50 weeks, while eight patients had disease progression. Five patients developed immune-related adverse events (irAEs).69

PREDICTIVE BIOMARKERS

In spite of the significant advancements in checkpoint inhibitors in the treatment of mUC and mRCC, the majority of patients are still not experiencing any benefits, while others have achieved long-term survivals. Attempts of discovering the biomarkers that are related to durable responses have commenced from investigating the PD-L1 expression, both on tumour cells and on tumour-infiltrating immune system cells. In cohort 2 of the IMvigor210 study, the patients with the PD-L1 expression ≥5% had the ORR of 26%, in comparison with the patients with <1% expression who responded in 8% of cases. The survival was also different in these two groups of patients, where the mOS was 11.4 months and 6.5 months in groups with ≥5% and <1%, respectively.17

In CheckMate-032 trial, patients treated with nivolumab had the mOS of 16.2 months versus 9.9 months in patients with the PD-L1 expression lower or higher than 1%, respectively.42 70 Similar results were observed in patients treated with durvalumab. In this study, the cut-off value for the PD-L1 positivity was significantly higher than in other trials. Patients with the PD-L1 expression higher
than 25% reached a response of 46.4% versus 0% in patients with the expression lower than the quoted cut-off.41 However, in both groups, survival benefits were present.41

Patients treated with pembrolizumab in the KEYNOTE-045 had shorter survival in the group with the expression ≥10% CPS, although pembrolizumab did result in benefits in both groups.37 Similarly, in the CheckMate-025 trial, nivolumab had longer survival versus everolimus, irrespective of the PD-L1 expression, whereas survivals were shorter in groups expressing ≥1%. This merely points to the worse prognosis of these patients and not to the response prediction to nivolumab.56 The heterogeneity of the results and a number of unanswered questions suggest that the PD-L1 expression is not a reliable prediction marker. Different companies use different cut-off values, antibodies, as well as different counting methods of the complete positivity of tumour and immune cells (CPS), or immune cells only as is the case in atezolizumab trials.17 This is the reason why a harmonisation of the PD-L1 status assessment, as well as a uniform assessment methodology in studies with different checkpoint inhibitors is of great necessity.

In the quest to define more appropriate biomarkers, the investigators of IMVigor210 trial have done the analysis of different subtypes of UC, as well as mutational load and T-cell infiltrate level.17 32 70 Although patients with basal subtype of UC had the highest expression rate of PD-L1, the best response to atezolizumab in cohort 2 had patients with luminal cluster II subtype.17 Similar results were reported for the cohort 1, however, with no statistical significance. Besides that, patients in cohort 1 have longer survival if they belong to luminal II and basal II (cluster IV) subtype.70 However, patients treated with nivolumab (CheckMate-275) had the best response with basal I subtype. It is quite clear that heterogeneity of results exists in different UC subtypes when considering different trials.

Tumours with higher mutational load have better response to checkpoint inhibitors. The patients with mUC treated with atezolizumab have better response if they have higher mutational burden.17 70 In cohort 1, the mutational load was a much better predictor of response than PD-L1 expression.70 Similar, density of CD8+ peritumour infiltrate was associated with better response of patients treated with atezolizumab.17

FUTURE DIRECTIONS AND CONCLUSION
The treatment of urological malignancies in the first-line urothelial cancer was not satisfactory. Patients who were eligible for optimal platinum-based chemotherapy had a median survival around 15 months in the metastatic disease setting. For platinum ineligible patients, survival was below 10 months. The results of the atezolizumab trials, as well as other PD-1/PD-L1 antibodies, have brought up hope for patients with mUC. The final results of phase III trials with pembrolizumab and atezolizumab in patients who progressed after cisplatinum-based treatment as well as cisplatinum–ineligible ones are expected.33 34 37 39 The results of avelumab in maintenance treatment,47 as well as combination of durvalumab and tremelimumab46 will give the answer about the position of these drugs in treatment of patients with mUC. Checkpoint inhibitors are also being assessed in adjuvant treatment of muscle-invasive35 and non-invasive UC.40

Nivolumab is already the standard of second-line treatment for mRCC.16 We expect the results with VEGF inhibitors in the first-line treatment, which might become a new standard of care.53

Since not all the patients respond to checkpoint inhibitors treatment, there is still a need to determine the adequate biomarkers for response prediction. Harmonisation of PD-L1 testing is one of the most important future issues to be resolved since immunohistochemistry method is inexpensive and readily available. Discovering new and easy to use biomarkers, better definitions of radiological response as well as the results of new trials will give the answer in the future, whether and to which extent will checkpoint inhibitors become the new state-of-the-art treatment of urological malignancies.

Contributors LSP wrote the manuscript and collected the data. GM-B collected the data. MP collected the data and made the tables.

Competing interests None declared.

Provenance and peer review Commissioned; externally peer reviewed.

Open Access This is an Open Access article distributed in accordance with the Creative Commons Attribution Non Commercial (CC BY-NC 4.0) license, which permits others to distribute, remix, adapt, build upon this work non-commercially, and license their derivative works on different terms, provided the original work is properly cited and the use is non-commercial. See: http://creativecommons.org/licenses/by-nc/4.0/

© European Society for Medical Oncology (unless otherwise stated in the text of the article) 2017. All rights reserved. No commercial use is permitted unless otherwise expressly granted.

REFERENCES


23. ClinicalTrials.gov Identifier: NCT02787005 Phase II trial ofPembrolizumab (MK-3475) in subjects with metastatic Castration-Resistant prostate Cancer (mCRPC) Previously treated with chemotherapy (KEYNOTE-199).


31. ClinicalTrials.gov Identifier: NCT02808766 A phase III, Multicenter, Randomized, Placebo-Controlled, Double-Blind Study of Atezolizumab (Anti-PD-L1 antibody) in combination with Gemcitabine/Carboplatin Versus Gemcitabine/Carboplatin alone in patients with locally Advanced or metastatic urothelial carcinoma who are ineligible for Cisplatin-Based therapy (IMvigor130).


33. ClinicalTrials.gov Identifier: NCT02853664 A phase III, Randomized, Controlled clinical trial of Pembrolizumab with or without Dabrafenib and Trametinib for the treatment of patients with BRAF-mutated metastatic melanoma.

34. ClinicalTrials.gov Identifier: NCT02853305 A phase III Randomized, Controlled clinical trial of Pembrolizumab with or without Platinum-Based Combination Chemotherapy Versus Chemotherapy in patients with advanced non-small cell lung Cancer.

35. ClinicalTrials.gov Identifier: NCT02625961 A phase II clinical trial to Study the Efficacy and Safety of Pembrolizumab (MK-3475) in subjects with high risk Non-muscle invasive bladder Cancer (NMIBC) Unre sponsive to Bacillus Calmette-Guerin (BCG) Therapy.


38. Galsky MD, Retz M, Stiefker-Radtke AO, et al. Efficacy and safety of nivolumab monotherapy in metastatic urothelial cancer (mUC) who have received previous treatment: results from the phase II CheckMate 275 study ESMO. J Clin Oncol 2016;LB.A31.


41. ClinicalTrials.gov Identifier: NCT02516241 A phase III, Randomized, Open-Label, Controlled, Multi-Center, Global Study of First-Line MED4736 Monotherapy and MED4736 in Combination with Tremelimumab Versus Standard of Care Chemotherapy in Patients with Unresectable Stage IV Urothelial Cancer.

42. ClinicalTrials.gov Identifier: NCT02603432 A phase 3, Multicenter, Multinational, Randomized, Open-label, Parallel-arm study of Avelumab (msb001781c) Plus best supportive care Versus Best supportive care alone as A Maintenance treatment in patients with locally Advanced or metastatic urothelial Cancer whose disease did not Progress after Completion of First-line Platinum-containing chemotherapy.


Checkpoint inhibitors in the treatment of urological malignancies

Lazar S Popovic, Gorana Matovina-Brko and Maja Popovic

ESMO Open 2017 2:
doi: 10.1136/esmoopen-2017-000165

Updated information and services can be found at:
http://esmoopen.bmj.com/content/2/2/e000165

These include:

References

This article cites 45 articles, 13 of which you can access for free at:
http://esmoopen.bmj.com/content/2/2/e000165#BIBL

Open Access

This is an Open Access article distributed in accordance with the Creative Commons Attribution Non Commercial (CC BY-NC 4.0) license, which permits others to distribute, remix, adapt, build upon this work non-commercially, and license their derivative works on different terms, provided the original work is properly cited and the use is non-commercial. See: http://creativecommons.org/licenses/by-nc/4.0/

Email alerting service

Receive free email alerts when new articles cite this article. Sign up in the box at the top right corner of the online article.

Topic Collections

Articles on similar topics can be found in the following collections

Open access (163)

Notes

To request permissions go to:
http://group.bmj.com/group/rights-licensing/permissions

To order reprints go to:
http://journals.bmj.com/cgi/reprintform

To subscribe to BMJ go to:
http://group.bmj.com/subscribe/