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Role of immunotherapy in bacillus Calmette-Guérin-unresponsive non-muscle invasive bladder cancer

Neelam Mukherjee¹, Robert S. Svatek¹, and Ahmed M. Mansour^{1,*}

¹Department of Urology, University of Texas Health Science Center at San Antonio, San Antonio, TX 78229, USA.

Abstract

Intravesical instillation of live attenuated *Bacillus Calmette–Guérin* (BCG) is the gold standard for patients with intermediate- and high-risk non-muscle-invasive bladder cancer (NMIBC). BCG-failures include a heterogeneous population of patients who share a designation of disease recurrence or progression following BCG and include patients with complete unresponsiveness to BCG, patients who respond initially but develop relapse and, in some cases, patients who are intolerant to BCG due to side effects. Given the efficacy and relatively rapid approval of several monoclonal antibodies against PD-L1 or PD-1 for advanced and metastatic bladder cancer, the role of these checkpoint inhibitors in BCG-relapsing disease at various disease stages is under consideration. Data supporting a role for immune checkpoint inhibitors is largely theoretical with limited supportive data from animal models and from clinical evidence of increased PD-L1 expression in BCG-unresponsive tumors. Current trials in BCG-unresponsive disease are underway and expected to provide insight regarding these concepts.

Defining BCG-Failures in NMIBC:

The prototypical treatment course for BCG is based on the original SWOG trial including a 6-week induction course followed by three weekly instillations at 3, 6, 12, 18, 30, and 36 months [1]. These three weekly instillations are often referred to as maintenance cycles and, therefore, complete treatment includes one induction course and 7 maintenance cycles. BCG failures are either (1) persistent growth of tumors that were present at the time of initiating BCG or (2) new tumors that were not present at the start of BCG. Patients experiencing disease relapse after receiving any amount BCG represent a heterogeneous population with varying amounts of risk that has been categorized according to the timing of relapse and amount of prior BCG before relapse. Some patients are intolerant to BCG and may not have been able to receive adequate dosing due to severe local side effects while other patients truly failed BCG [2–4]. Therefore, for the purpose of standardization, BCG-relapsing disease is defined as recurrence of high grade disease after achieving a disease-free state at 6 months provided that adequate BCG was administered [5].

*Correspondence: Ahmed M. Mansour, MD, Assistant Professor, Department of Urology, The University of Texas Health Science Center San Antonio, 7703 Floyd Curl Dr. MC 7845, San Antonio, Tx 78229, Phone: 210-567-0115, Fax: 210-567-6868, mansour@uthscsa.edu.

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BCG unresponsive disease is the latest reiteration of defining BCG failures and was the term adopted by the Genitourinary American Society of Clinical Oncology (GU ASCO) Group and the International Bladder Cancer Group (IBCG) to identify patients in whom further BCG is not indicated [3, 6]. BCG unresponsive disease denotes a group of patients with a particularly poor prognosis and includes patients who are refractory to BCG or those who relapse within 6 months of the last BCG exposure [7]. These particular definitions are relatively new and may not be used in clinical trials conducted prior to 2015. BCG-refractory implies no response to BCG and is defined as persistent high-grade bladder cancer at 6 months after the start of induction therapy or tumors that have progressed by grade or stage at 3 months after the start of BCG induction therapy [5]. Inherently, then, these definitions are linked to the adequacy of the tumor excision prior to initiation of adjuvant BCG. Incomplete resection or non-detection of a high-grade T1 tumor, for example, may be best described as a surgical deficiency but could erroneously lead to the designation of a patient as “BCG-unresponsive”.

To meet these definitions, how much BCG is required to be delivered? Currently, no designation for dose per instillation is unanimously adopted. However, from a clinical trial design perspective, adequacy has recently been designated as the receipt of at least 5 of 6 intended instillations for induction and at least 2 instillations of maintenance BCG [3, 6] or a 2nd re-induction course of at least 5 instillations [5, 6, 8].

Current management of BCG–unresponsive disease:

Many patients are not truly BCG-unresponsive but instead have received inadequate BCG (see above). In truly BCG-unresponsive patients, the current “standard of care” could be considered a radical cystectomy. An alternative to cystectomy – either due to patient refusal or due to medical judgement based on patient’s comorbid conditions, valrubicin is the only Food and Drug Administration (FDA) approved intravesical agent available, although it is approved only for BCG-refractory CIS. Importantly, the long-term efficacy of valrubicin in this setting is modest with a disease-free response rate of only 16% at 12 months [7]. The current practice patterns for management of patients with BCG-unresponsive are not well documented. Based on discussions with bladder cancer experts in the U.S., patients with truly BCG-unresponsive disease currently undergo diverse management approaches, many of which are off-trial and include re-induction BCG +/- additional intravesical agents, intravesical chemotherapy either alone or in combination with other chemotherapies, alternative intravesical immunotherapies, and in some cases systemic therapy. Although valrubicin is only FDA approved for CIS, it has been used to treat BCG-unresponsive Ta-T1 disease. Generally, an increasing number of patients are managed in clinical trials.

PD-L1/PD-1 checkpoint inhibitors – mechanisms and current approvals

The two checkpoint proteins which gained critical importance in the context of bladder cancer are (programmed death ligand-1 [PD-L1] and programmed cell death protein-1 [PD-1] [9, 10]. T lymphocytes (T cells) can differentiate between normal healthy cells and infected or cancerous cells through antigen-specific T cell receptors and associated co-activation and co-inhibitory receptors expressed by the T-cells. The current theory regarding

mechanism of PD-L1/PD-1 axis proposes that PD-L1 expressed on the surface of tumors binds to PD-1 expressed on T cells and transmits an inhibitory signal to the T cell. Thus these ligand/receptor interaction limits the effector arm of immune system and keeps their response ‘in check’ thereby preventing auto immunity and tissue destruction. Thus, they are referred to as the checkpoint proteins. Tumor cells can sometimes evade the immune response of the T cells essentially by simulating the signals of the normal and healthy cells [11]. As a result, the immune system can no longer restrict the growth of these malignant cells allowing the tumor to grow and proliferate. Checkpoint inhibitors modulate the activity of these checkpoint proteins, allowing them to “release the brakes” for effective immune response against the malignant cells [12].

PD-1/PD-L1 pathway functions by activating a series of events [11]. The pathway includes the receptor PD-1 and its 2 ligands, PD-1 ligand 1 (PD-L1, B7-H1, and CD274), and PD-1 ligand 2 (PD-L2, B7-DC, and CD273), which are cell surface glycoproteins belonging to the B7 family. PD-L1 is expressed on a variety of cells including human antigen-presenting cells, T cells, natural killer cells, stem cells and tumor cells [13]. However, the expression of PD-L2 is limited to a restricted population of cells and very limited information exists on its role in tumor biology. Binding of PD-L1 and PD-L2 to PD-1 (expressed on activated and exhausted T cells, macrophages, dendritic cells, and B cells) [14] results in the phosphorylation of the immunoreceptor tyrosine-based switch and recruitment of phosphatases SHP-1 and SHP-2 [15] thereby regulating T-cell antigen receptor, cytokine production, T-cell activation, and target cell lysis. PD-L1 has also been shown to regulate several genes involved in tumor cell proliferation such as mammalian target of rapamycin (mTOR) and autophagy pathways [16]. It also regulates functional tumor initiating cell (TIC) generation and canonical TIC gene expression [17].

Levels of PD-L1 expression have been shown to be associated with bladder cancer outcome. Patients who have tumors with higher levels of PD-L1 have higher recurrence and poorer survival [18–20]. However, since patients with tumors expressing low PD-L1 can respond to checkpoint inhibition and because some patients with high PD-L1 do not respond, this represents an incomplete understanding of the mechanisms involved in response to these agents. Check point inhibitors seem to be very effective in cancers with very high mutational rate [21] such as bladder cancer, since these cancers elicit very strong T cell responses.

Approved Checkpoint Inhibitors:

Atezolizumab (TECENTRIQ®, Genentech) was the first PD-L1 inhibitor approved for patients with locally advanced or metastatic bladder cancer who progressed on or after platinum-based chemotherapy or have progressed within 12 months of neoadjuvant or adjuvant treatment with a platinum-containing chemotherapy. It was given accelerated approval by the FDA in May 2016 [9, 22] but later the approval was extended in April, 2017 to treatment-naïve patients with locally advanced or metastatic disease and who are not eligible for cisplatin-containing therapy. However, Tecentriq was approved based on its phase 2 data and unfortunately it failed its Phase 3 trial and could not prolong patients’ lives raising the concern whether FDA will rescind its approval of the drug.

Other approved PD-L1 targeting antibodies include Avelumab (BAVENCIO®, EMD Serono, Inc.) [23] and Durvalumab (Imfinzi™, AstraZeneca UK Limited) [10]. Both Avelumab and Durvalumab received approval from FDA in May for patients with locally advanced or metastatic bladder cancer and have progressed during or after platinum-containing chemotherapy or within 12 months of neoadjuvant or adjuvant chemotherapy.

Nivolumab (Opdivo®, Bristol-Myers Squibb) is a fully human immunoglobulin monoclonal antibody that targets the PD-1 receptor and was granted accelerated approval by FDA on February 2nd, 2017 for patients with locally advanced or metastatic urothelial carcinoma who have progressed during or following platinum-containing chemotherapy or have progressed within 12 months of neoadjuvant or adjuvant treatment with a platinum-containing chemotherapy [24].

Pembrolizumab (Keytruda®, Merck & Co) is a humanized monoclonal antibody [25, 26] that targets the PD-1 receptor and it received regular approval very recently on May 18th, 2017 by FDA for patients with locally advanced or metastatic bladder cancer who have progressed during or after platinum-containing chemotherapy or within 12 months of neoadjuvant or adjuvant chemotherapy. It also received accelerated approval on 17th April, 2017 for patients with locally advanced or metastatic bladder cancer who are not eligible for cisplatin-therapy.

Rationale for using immune therapy in BCG-unresponsive disease

With the success of checkpoint immunotherapy in metastatic bladder cancer, combination therapy of checkpoint inhibitors in BCG unresponsive bladder cancer has been considered. Importantly, PD-L1 expression has been linked with increased resistance to BCG therapy and BCG granulomas in BCG-unresponsive patients exhibit high levels of PD-L1 expression [19]. This suggests that PD-L1 may help the progression of bladder tumor into deeper structures of the bladder by suppressing T cell immune response and subsequent BCG failure. As a result, use of PDL1 inhibitors in BCG unresponsive bladder cancer is a plausible strategy for testing.

CpG oligodeoxynucleotides (ODNs) are immune activating 20-bp oligonucleotides containing a cytosine triphosphate deoxynucleotide (“C”) followed by a guanine triphosphate deoxynucleotide (“G”). They are labelled as pathogen-associated molecular pattern motifs (PAMPs) because they are highly frequent in microbial genomes (such as BCG DNA) in contrast to the vertebrate genomes [27]. CpG ODNs have been shown to have therapeutic efficacy in MB49 bladder tumor murine model. They mediated tumor-specific memory T-cell response preventing tumor development whereas BCG was not found to be very effective in this model [28]. It was also shown that even though there were T cells present in the MB49 tumor model they could not prevent tumor progression. One of the possible explanations can be that T cell inhibitory signals like PD-L1 and CTLA-4 were upregulated and prevented an adequate immune response.

Current ongoing immune-therapy clinical trials for BCG-unresponsive disease:

I -Checkpoint inhibitors clinical trials:

I.1. Single agent checkpoint inhibitors

I.1.a Atezolizumab: The ongoing phase II trial sponsored by the NCI is investigating Atezolizumab as a single agent in BCG unresponsive patients (NCT02844816). NCT02451423 is another currently recruiting phase II trial investigating neoadjuvant Atezolizumab prior to radical cystectomy in patients with BCG-unresponsive NMIBC

I.1.B Durvalumab: Currently recruiting phase II study investigating Durvalumab (Medi4736) for BCG unresponsive CIS of the Bladder (NCT02901548) by looking into complete response rates at 6 months and 24 months

I.1.C Pembrolizumab: NCT02625961 trial is single arm phase II trial. It is currently investigating Pembrolizumab in patients with high risk BCG-unresponsive NMIBC who are ineligible or refuse radical cystectomy

I.2. Combination therapy with BCG

I.2.a Atezolizumab +/- BCG: Phase Ib/II study is currently ongoing assessing the safety, tolerability, pharmacokinetics, immunogenicity and preliminary anti-tumor activity of atezolizumab alone and in combination with intravesical BCG in high-risk NMIBC participants. Primary outcome is the percentage of patients with adverse events (NCT02792192).

I.2.b Pembrolizumab + BCG: NCT02324582 is an ongoing single center Phase I safety and efficacy study of Pembrolizumab used in combination with CG instillation for management of patients with high risk NMIBC. Patients are given 6 cycles (21 days) of IV pembrolizumab in combination with BCG 6-weekly course. BCG treatment begins on Day 1 of cycle 3 of pembrolizumab.

II -Vaccine therapy clinical trials

II.1. BCG +/- PANVAC—PANVAC is a vector-based vaccine that contains tumor associated antigens and induce a CD4 and CD8 antigen specific response [29]. A prospective randomized phase II study is ongoing to determine the efficacy of intravesical BCG in combination with subcutaneous PANVAC versus BCG alone in high grade BCG-unresponsive bladder cancer (NCT02015104).

II.2 ALT-803—Alt-803 is a recombinant fusion protein with enhanced IL-15 activity which is important for the development and activation of natural killer and CD-8 cells [30]. Ongoing current trial of Phase Ib/II, multicenter study of intravesical BCG plus ALT-803 in high risk NMIBC. Efficacy is evaluated by assessing the complete response rate over a 4-year period. (NCT02138734). For BCG-unresponsive patients, a phase Ib/II, multi-center

trial of IV ALT-801 combined with IV gemcitabine (NCT01625260) in patients who refuse or are unfit for radical cystectomy.

III -Gene therapy clinical trials

III.1 rAd-IFN α /Syn3 (Instiladrin)—rAd-IFN non-replicating adenovirus vector harboring the human IFN α 2b gene. When combined with the excipient Syn3, intravesical administration of the rAd-IFN results in transduction of the virus into the epithelial cell lining in the bladder. The IFN α 2b gene is incorporated into the cellular DNA resulting in the synthesis and expression of large amount of IFN α 2b protein allowing for high and durable IFN α in urine [31]

Most recently, results of phase II, multicenter, study were reported (NCT01687244). 43 patients with high grade BCG-refractory or relapsed NMIBC received intravesical rAd-IFN α /Syn3. The primary end was 12-month high-grade recurrence-free survival. Durable responses were noted in 35% of patients, which represents more than a three-fold improvement in recurrence-free survival compared to Valrubicin, the only agent currently approved for this indication [32]. Expanding on these results, an ongoing Phase III trial (NCT02773849) with estimated enrollment of 135 patients is currently recruiting.

III.2 CG-0700—CG0070 is an oncolytic adenovirus that expresses the immune stimulatory cytokine GM-CSF [33]. It is being administered intravesically as a single-arm intervention in an open-label, phase III study (NCT02365818). The ongoing trial recently reported interim analysis, documenting overall 47% complete response rate at 6 months for all patients and 50% for patients with CIS [34].

Summary and Future directions

We are witnessing an important era in the management of bladder cancer; the rapid progress in cancer immunotherapy in the past few years is reshaping the field. The success story of how immunotherapy and personalized medicine have affected the management of melanoma is definitely intriguing. A plethora of clinical trials are currently ongoing propelled by the rising interest in the field, however, this requires consolidation of efforts for successful completion of these trials. Standardized new definitions for BCG unresponsiveness needs to be adopted even in already started trials.

A variety of rational combinations of immunotherapy and targeted therapy agents are worth investigating. New trial design should consider multi-arm clinical trials comparing several treatments against a common group and selecting promising treatments to continue; this will allow for significant cost reduction of the trials and increase the chances of their completion.

Finally, refinements in PD1 and PDL1 expression assays will allow for personalization of management. Mutational load and genetic instability will play an important role as a predictive marker for immune-response and may aid in directing patients to immunotherapy.

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